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DORMANCY PERIODS OF PHILIPPINE SEED BOARD RICE VARIETIES [1]

R. C. Dumlao, P. B. Escuro, and M. B. Parker

WITH TWO TEXT FIGURES

Weekly germination tests of seven upland and eight lowland Seed Board rice varieties were conducted starting immediately after harvest until germination reached about 90 per cent to determine the dormancy or rest period of the grains. Dormancy was arbitrarily considered to have ended when germination reached 80 per cent. In general, the early varieties had a shorter dormancy period than the late ones. The highly significant correlation coefficient between the maturity and dormancy period ($r = 0.672$) indicates a positive association between these characters. Among the upland varieties, Pinulot had the longest dormancy (7 weeks), followed by Azucena and Magsanaya (6 weeks), Nagdami (5 weeks), Palawan and Fortuna (4 weeks), and Mangarez (3 weeks). Among the lowland varieties, Tjahaja had the longest (10 weeks), followed by B-E-3 and Intan (9 weeks), Raminad Str. 3 and Seraup Kechil 36 Str. 482 (8 weeks), and BPI (FB)-76, Tjere Mas, and Peta (7 weeks).

Freshly harvested seeds of certain rice varieties either do not germinate at once or give a low percentage of germination (Pan, 1936; Buenaventura, 1956). These varieties need a rest or dormancy period before a high percentage of germination can be obtained.

A reasonably short dormancy period is advantageous. The absence of dormancy in a variety may cause maturing grains to sprout during wet weather. On the other hand, too long dormancy prevents the use of newly harvested grains for planting. It also causes delay in the analysis of seed samples submitted to the Certifying Agency.

Dormancy has been attributed to a number of causes. It is claimed to be a natural follow-up of the maturation process (Wilson, 1948). It is exhibited by seeds possessing one or more of the following: (1) a thick or hard seed coat that prevents the absorption of water, (2) a seed coat that interferes with the penetration of oxygen, (3) an immature embryo, and (4) an embryo which is apparently mature but which undergoes changes before germination (Crocker,

[1] Central Experiment Station Contribution No. 2050.

1916, as cited by Wolf and Kipps, 1953). A chemical inhibitory to germination of seeds in the chaff of some desert seeds has been reported (Bonner and Galston, 1952). The degree of maturity also affects inversely the length of dormancy in rice (Jones, 1926; Lord, 1929; Umali, Marquez, and Lantican, 1957[2]). Dormancy of barley seeds which mature during the rainy season has been attributed to genetic and environmental factors at the time of growth and maturation (Crocker and Barton, 1953). Realizing that varietal differences exist, this work was conducted to determine the dormancy periods of Seed Board rice varieties.

MATERIALS AND METHODS

Foundation seeds of seven upland and eight lowland Seed Board rice varieties harvested at approximately the same stage of maturity were used in this study. Germination tests were conducted at weekly intervals until germination was about 90 per cent. One hundred well-filled grains of each variety were spread uniformly in each of three Petri dishes provided with moist tissue paper and incubated under room conditions. Percentages of germination were determined one week later. Except for the first germination test in which newly harvested grains were used, the bulked seeds of each variety dried to about 13 per cent moisture content and stored in cans under ordinary room conditions served as the common source of seed for the tests. The dormancy period of a variety was arbitrarily considered to have ended when germination reached 80 per cent (Chandraratna, Fernando, and Wattegedera, 1952; Buenaventura, 1956).

RESULTS AND DISCUSSION

Based on the assumption that rice dormancy has ended when germination has reached 80 per cent, the upland varieties had rest periods ranging from three to seven weeks (table 1 and fig. 1). Pinulot had the longest dormancy (7 weeks), followed by Azucena and Magsanaya (6 weeks), Nagdami (5 weeks), Palawan and Fortuna (4 weeks), and Mangarez (3 weeks). On the other hand, the lowland varieties had longer rest periods ranging from seven to ten weeks (table 1 and fig. 2). BPI (FB)-76, Tjere Mas, and Peta were dormant for seven weeks, Raminad Strain 3 and Seraup Kechil 36 Strain 482 for eight weeks, B-E-3 and Intan for nine weeks, and Tjahaja for 10 weeks.

[2] D L. Umali, L. D. Marquez, and R. M. Lantican, "Influence of degree of maturity on dormancy of the rice grain. 1957. pp. 1-5. (Mimeo.).

TABLE 1.—*Percentage of germination determined at weekly intervals, standard errors, dormancy periods and maturity of Philippine Seed Board rice varieties*

VARIETY	WEEKS AFTER HARVEST											EST. DORMANCY	MATURITY
	0	1	2	3	4	5	6	7	8	9	10		
Upland												*weeks*	
Mangarez	6.7±0.20	11.3±3.38	56.7±2.03	94.3±1.86	—	—	—	—	—	—	—	3	19.1
Fortuna	—	30.0±4.62	77.7±2.20	76.7±4.26	91.3±2.03	—	—	—	—	—	—	4	19.8
Palawan	2.0±0.57	1.3±0.34	5.0±1.73	72.7±1.46	91.0±3.05	—	—	—	—	—	—	4	19.2
Nagdami	—	5.7±0.88	16.5±0.40	50.0±2.53	58.0±0.57	85.3±2.33	—	—	—	—	—	5	19.5
Magsanaya	—	—	—	11.3±1.20	58.7±2.85	77.3±2.67	86.7±2.33	—	—	—	—	6	18.0
Azucena	16.0±2.62	17.0±1.16	40.7±2.33	50.3±3.29	66.7±1.77	66.0±7.21	80.3±1.45	—	—	—	—	6	18.7
Pinulot	—	10.7±2.33	17.3±0.34	21.7±3.76	38.7±2.03	56.0±3.61	76.7±1.33	86.0±2.52	—	—	—	7	19.1
Lowland													
BPI (FB)-76	—	0.7±0.34	3.3±0.88	31.7±1.76	42.0±1.76	59.3±1.47	68.7±3.18	87.7±1.45	—	—	—	7	21.8
Tjere Mas	0	11.3±2.33	21.3±2.33	37.0±2.00	42.0±5.81	67.7±2.60	71.3±0.34	89.0±1.53	—	—	—	7	20.1
Peta	0	0	15.7±0.89	16.3±1.33	29.7±2.33	43.7±8.00	71.3±3.67	93.0±1.00	—	—	—	7	21.1
Raminad Str. 3	0	1.7±0.67	4.0±0.57	24.7±0.88	32.3±1.20	39.7±3.18	47.3±7.22	50.0±4.51	80.3±0.88	—	—	8	24.4
SK. 36 Str. 482	0	11.0±2.67	15.0±1.15	34.0±1.00	43.3±4.10	57.3±4.49	59.7±1.86	71.7±1.45	86.3±1.45	—	—	8	25.1
B-E-3	0	1.7±0.67	5.5±0.50	18.7±0.67	28.5±3.51	30.5±0.50	51.0±6.01	61.3±0.88	77.3±0.34	91.0±3.60	—	9	22.2
Intan	0	0	1.0±1.00	0.3±0.34	24.7±3.29	41.3±9.89	72.0±3.05	74.7±2.73	75.3±0.89	82.7±3.39	—	9	21.5
Tjahaja	0	0	0.7±0.67	0.3±0.34	9.7±0.88	11.0±2.64	26.3±3.53	43.0±3.51	43.7±4.37	78.0±3.22	82.0±1.00	10	22.8

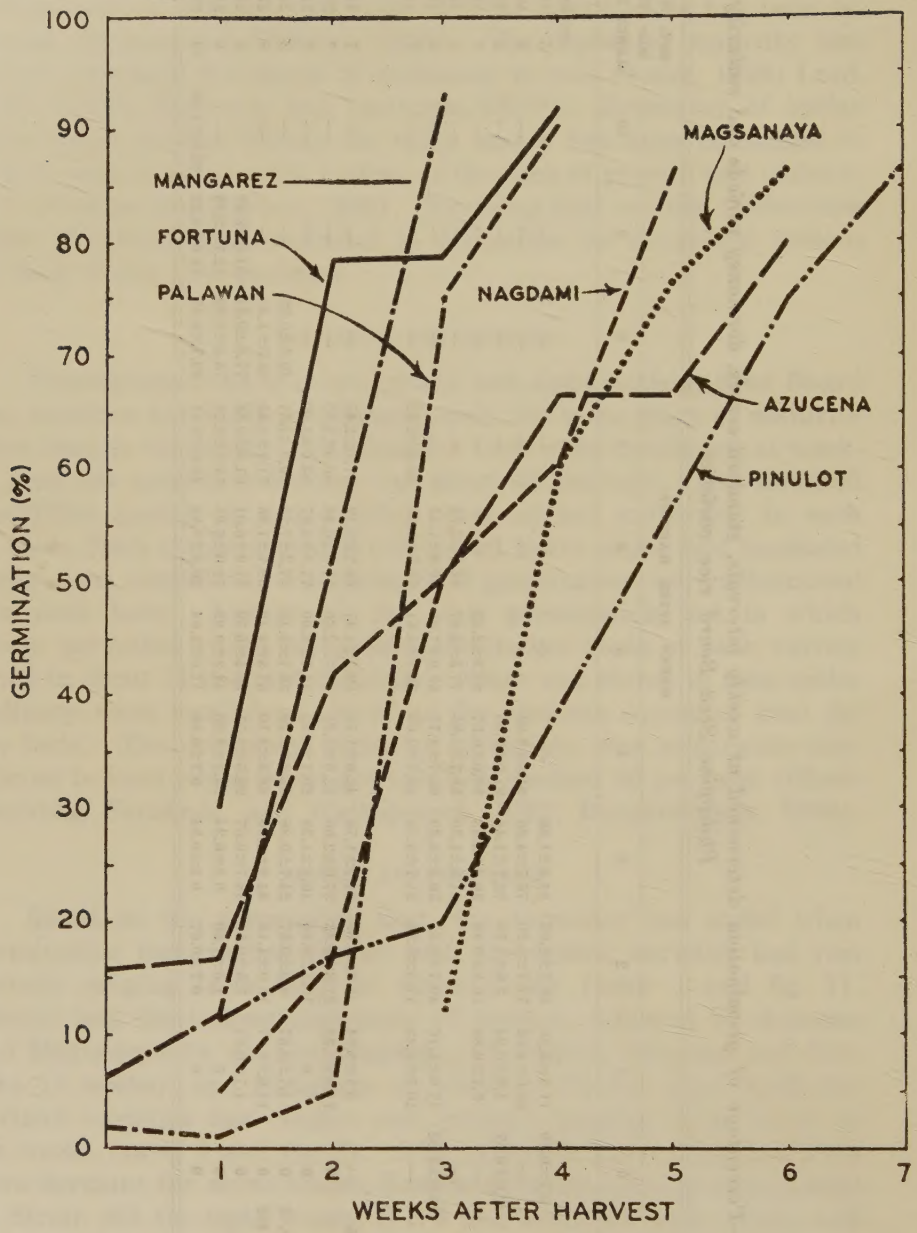

Fig. 1.—Trend of germination of Seed Board upland varieties determined at weekly intervals after harvest.

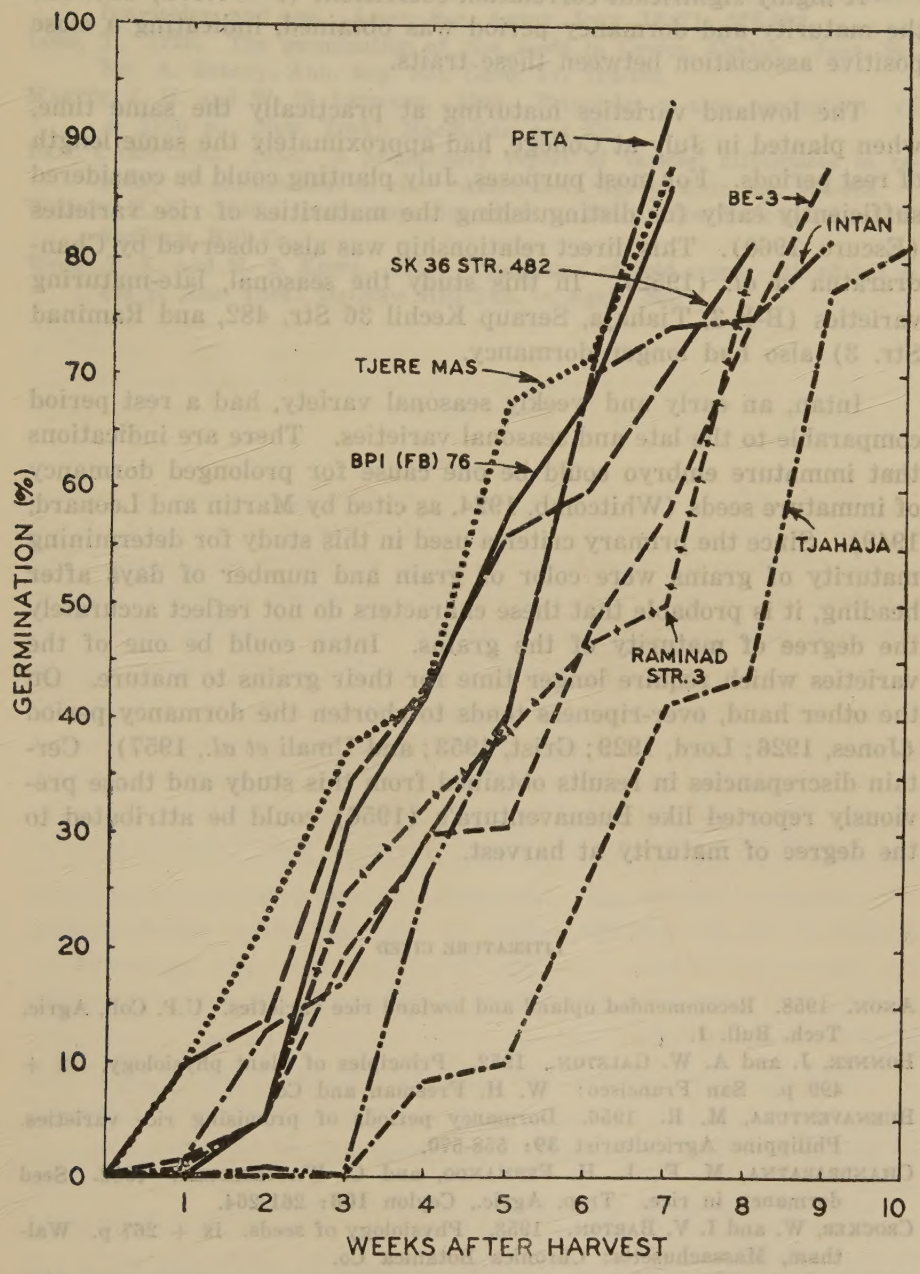

Fig. 2.—Trend of germination of Seed Board lowland varieties determined at weekly intervals after harvest.

A highly significant correlation coefficient ($r = 0.672$) between the maturity and dormancy period was obtained, indicating a close positive association between these traits.

The lowland varieties maturing at practically the same time, when planted in July at College, had approximately the same length of rest periods. For most purposes, July planting could be considered sufficiently early for distinguishing the maturities of rice varieties (Escuro, 1960). This direct relationship was also observed by Chandraratna *et al.* (1952). In this study the seasonal, late-maturing varieties (B-E-3, Tjahaja, Seraup Kechil 36 Str. 482, and Raminad Str. 3) also had longer dormancy.

Intan, an early and weekly seasonal variety, had a rest period comparable to the late and seasonal varieties. There are indications that immature embryo could be one cause for prolonged dormancy of immature seeds (Whitcomb, 1924, as cited by Martin and Leonard, 1949). Since the primary criteria used in this study for determining maturity of grains were color of grain and number of days after heading, it is probable that these characters do not reflect accurately the degree of maturity of the grains. Intan could be one of the varieties which require longer time for their grains to mature. On the other hand, over-ripeness tends to shorten the dormancy period (Jones, 1926; Lord, 1929; Grist, 1953; and Umali *et al.*, 1957). Certain discrepancies in results obtained from this study and those previously reported like Buenaventura's (1956) could be attributed to the degree of maturity at harvest.

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THE RELATIVE EFFICIENCY OF THE 4 × 4 LATIN SQUARE FOR TESTING GRAIN AND STOVER YIELDS OF CORN[1]

I. S. SANTOS and B. L. LIT

For comparing grain yields, the results of this study suggest that it would probably be best to put the blocks of the simple randomized complete block experiment running perpendicular to the Pili Avenue. When dealing with a small number of varieties or entries and in the absence of any definite information about the presence and direction of any soil-fertility gradient, the use of the Latin square design is hereby suggested for use in the Central Experiment Station fields. The 4 x 4 Latin square design was superior to the simple randomized complete-block design on the average, in comparing entries with respect to grain yield and stover yield.

When only a few entries are to be tested, the use of experimental designs that provide for precision greater than that of the simple randomized complete-block design should be considered. For advanced performance tests involving only three to eight entries, the Latin square, a design perfected by Fisher, appears suitable.

The term *Latin square* was first used by Euler in his study of the different possible square arrangements. This term was subsequently introduced into agricultural science to designate a square chosen at random out of the total number of these possible arrangements. The value of the Latin square design as a means of eliminating variation due to soil fertility differences was recognized early.

Snedecor (1937)[2] said that under suitable conditions, the Latin square is, on the average, about 20 per cent more efficient than the randomized blocks. He also stated that the Latin square is especially well adapted to the field trial of usually not more than eight treatments or varieties.

Cochran and Cox (1950)[3] asserted that the Latin square provides more opportunity than randomized blocks for the reduction of errors by skillful planning. In Latin square field experiments,

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[1] Central Experiment Station Contribution No. 2051.

[2] G. W. Snedecor, "Statistical Methods," 4th ed., Iowa State College Press, Ames, Iowa, 1937, p. 271.

[3] W. G. Cochran and G. M. Cox, "Experimental Designs," John Wiley and Sons, Inc., New York, 1950, p. 103.

the plots are usually laid out in a square formation, thereby reducing as much as possible, variation due to heterogeneity and other factors.

Yates (1933)[4] reported that randomized block arrangements, being very flexible, are eminently suitable for many experiments. On the other hand, the Latin square where each treatment or entry occurs only once in each row and each column, may be expected on the average to eliminate variations due to soil heterogeneity more completely when the size of the square is not too large, since soil variation in two directions at right angles affect all treatments equally.

The field experiments reported in this paper were conducted from November, 1955 to October, 1956 at the Central Experiment Station, U.P. College of Agriculture.

MATERIALS AND METHODS

Three promising corn double crosses—(A103 x A107) (A111 x A113), (192 x A112) (A102 x A103), and (I17 x I18) (A111 x A113)—and College Yellow Flint, a high-yielding open-pollinated variety, were the entries used in this study.

The plantings were done on the fourth 50-meter wide hectare lot on the left side of the Pili Avenue (when facing the railroad tracks) between Mt. Makiling and the Manila Railroad tracks.

A 4 x 4 Latin square design was used for all plantings except in the first, which was inadvertently planted as a simple randomized complete-block experiment. However, the second planting was planted twice on the same day so that there were six Latin squares in all.

Each plot was five rows wide and nine hills long. Rows were one meter apart; hills in the rows, one-half meter apart. Five kernels were dropped to a hill but the hills were thinned to a maximum of two plants each.

For the 1955-56 dry season, plantings were made on November 10 and 23, and December 7, 1955. For the 1956 wet season, plantings were done on May 7 and 21, and June 13, 1956.

The ears were harvested clean of husks from all five rows of each plot and allowed to dry in a forced-heated-air dryer to about

[4] F. Yates, *Emp. Jour. Exp. Agr., 1,* 235-244 (1933).

14 per cent moisture content. Ears were weighed just before shelling. Immediately after shelling, the shelled grains were weighed. A shelled grain sample from each plot was tested for moisture content using the Universal Electronic Moisture Tester. Grain yield was expressed in cavans per hectare at 15 per cent moisture. The stand after thinning was used as the basis in converting plot yields into hectare yields. For each missing hill, the plot was assumed to have lost 0.9 of a hill; for each one-stalk hill, 0.3 of a hill.

The stalks and the leaves (including the husks) from all five rows of each plot were harvested and weighed for stover yield determination. Five stalks taken at random from each plot were weighed separately and dried in the dryer to "constant weight". The final weights of these samples were taken and moisture content of the stalks in the plot from which the sample was taken was computed. Stover yield was expressed in tons per hectare at 15 per cent moisture. As in grain yield, the stand after thinning was used in converting plot yields to hectare yields.

The method of computing the relative efficiency of the Latin square as compared to the randomized block design followed was that given by Cochran and Cox (1950).[5]

RESULTS AND DISCUSSION

Table 1 shows the percentage of efficiency of the 4 × 4 Latin square design as compared to the randomized complete-block design when dealing with grain yield, stover yield, and reaction to the downy mildew disease. The *c* columns include percentage figures on the efficiency of the 4 × 4 Latin square as compared to a simple randomized complete-block design with columns as blocks that might have been used instead of the Latin square. A similar explanation holds true for the *r* columns. The simple randomized block designs mentioned here are not those obtained by regrouping (using either the columns or rows to serve as blocks) the Latin square data but rather are those that could have been used (hypothetically) instead of the Latin square design.

Dry season, 1955-56. The Latin square design was superior to either randomized block design in the measurement of grain yield in the (A) planting of November 23, 1955, the mean comparative efficiency being 156.0 per cent (or a gain of 56.0 per cent on the

[5] Cochran and Cox, *op. cit.*, p. 112.

TABLE 1.—*Efficiency (per cent) of the 4 × 4 Latin square design as compared to the simple randomized complete block design*

PLANTINGS	GRAIN YIELD			STOVER YIELD			DOWNY MILDEW		
	c[a]	r[b]	Mean	c[a]	r[b]	Mean	c[a]	r[b]	Mean
Nov. 23, 1955(A)	143.0	169.0	156.0	132.2	171.1	151.6	c	c	—
Nov. 23, 1955(B)	77.3	94.9	86.1	171.6	121.4	146.5	c	c	—
Dec. 7, 1955	80.8	367.9	224.4	d	d	—	c	c	—
May 7, 1956	—	—	—	—	—	—	149.5	77.0	113.2
May 21, 1956	111.6	81.7	96.6	107.5	94.8	101.2	115.4	99.9	107.6
June 13, 1956	96.2	97.2	96.7	180.3	89.7	135.0	129.5	80.9	105.2
Mean	101.8	162.1	132.0	147.9	119.2	133.6	131.5	85.9	108.7

[a] As compared to a simple randomized complete-block design with columns as blocks.

[b] As compared to a simple randomized complete-block design with rows as blocks.

[c] Downy mildew percentages were very small or negligible.

[d] Stalks of two plots of (I17 × I18) (A111 × A113) were stolen before these could be harvested. Data on stover yield were analyzed as if taken from a completely randomized experiment.

average). In the (B) planting of November 23, 1955, however, the mean efficiency was only 86.1 per cent, indicating a loss of 13.9 per cent. In this (B) planting, it was obvious that it would have been better if a simple randomized complete-block design with columns as blocks was used instead of the Latin square. In the December 7, 1955 planting, a simple randomized complete-block design with rows as blocks would have been decidedly inferior to the Latin square.

With stover yield, the Latin square design was superior to both randomized block arrangements. The "trend" for stover yield was exactly opposite that of grain yield.

Wet season, 1956. For grain yield, the "trend" in the May 21, 1956 planting was exactly opposite that in the 1955-56 dry season. Were a randomized complete-block design used, it should have been one with the rows, instead of columns as blocks. In this planting, the Latin square still had a slight advantage over the simple randomized block design on the average. In the June 13, 1956 planting, it would not have mattered whether a Latin square design or a simple randomized complete-block design was used.

For stover yield, the same "trend" as was present in the 1955-56 dry season planting existed in the May 21 and June 13, 1956 plantings. In the June 13, 1956 planting, the Latin square had a gain

in efficiency on the average, but were a simple randomized block design used, it should have been one with rows as blocks.

In all the 1956 wet season plantings, the Latin square arrangement registered gains, on the average, in the measurement of downy mildew infection. It would have been better however, if a simple randomized complete-block design with rows as blocks were used, especially on the May 7 and June 13, 1956 plantings.

SUMMARY AND RECOMMENDATION

In all the Latin-square plantings, the *columns* ran perpendicular to the Pili Avenue; the *rows,* parallel to the Pili Avenue.

In all the dry-season plantings (except the November 10, 1955 planting), the soil-fertility gradient, as it affected grain yield, ran in the same direction as the Pili Avenue. In the May 21, 1956 planting, when downy mildew infection was considerable, soil variability as caused by fertility gradient was overruled by the variation in amount of downy mildew inoculum as affected by distance from the source. The main source of the downy mildew inoculum was a downy-mildew-infected corn planting at one end of the 50 $\times$ 200 meter lot in which the wet-season Latin square experiments were planted. In the June 13, 1956 planting it was either that the few plants on which the grain yields were based did not reflect the soil-fertility gradient or that the opposite effects of soil fertility gradient and of "downy-mildew gradient" counterbalanced each other.

If the explanation just given was true, then it would be difficult to explain why the "trend" for stover yield in the dry-season plantings was opposite that for grain yield. It is possible that soil-fertility gradient did not affect stover yields in the dry season and that the slight downy-mildew infection in that season had a "gradient" similar to that in the wet season of 1956.

For measuring grain yield, the means at the bottom row of table 1 indicated that the blocks of a simple randomized complete-block experiment should run perpendicular to the Pili Avenue. The situations for stover yield and downy mildew infection were similar, indicating a close relationship between them. It is therefore unwise to recommend how blocks should be laid out when dealing with stover yield and downy mildew infection. For downy mildew infection, the source or possible source of infection must be considered.

GENETIC ASPECTS OF THE INTERSPECIFIC CROSSES BETWEEN KENAF (HIBISCUS CANNABINUS L.) AND ROSELLE (HIBISCUS SABDARIFFA L.)[1]

CHARITO A. FUKUSAWA, FILOMENA F. CAMPOS, and JOSÉ M. CAPINPIN

WITH FOUR TEXT FIGURES

Hybrids were produced from crosses between *Hibiscus cannabinus* L. and *Hibiscus sabdariffa* L. No self and backcross progenies were obtained due to sterility exhibited by the F_1 hybrids. Agronomic and morpho-genetic comparisons of the F_1 hybrids and the parental species were observed and compared. Among the striking characters of the hybrids are increased height, delayed time of flowering, sterility and dense trichome condition. All other characters are either intermediate or similar to either parent.

Since its introduction, kenaf has revitalized the fiber industry in the Philippines. It is not only easy to grow but it also yields comparatively more fiber than jute.

According to Crane (1947), investigations in the United States and Cuba showed that greater yields of fiber per unit of land were obtained from the kenaf plant than from several other plants studied. But kenaf has one objectionable characteristic: trichomes, which produce an itchy effect, are found on almost every part of the plant. Their presence reduces the efficiency of the harvesters, thus making the harvest period the most expensive phase of kenaf production. Sison (1954) found that variety *vulgaris* College Selection F_7 has the most trichomes.

Roselle (*H. sabdariffa* L.) is a closely related member of the genus. It is used principally for its bast fiber and for its fruit, particularly the calyx which is made into jams.

Crane (1948) noted that the silky, soft and light-colored fiber obtained from roselle has practically the same chemical and physical properties as jute (*Corchorus capsularis* L.) fiber.

Mendiola (1956) also found that roselle fiber appears almost as white as abaca and shows a considerable degree of brilliancy. It compares favorably in strength with various cordage fibers.

[1] Central Experiment Station Contribution No. 2052. Research Project 1362.

The roselle plant has also one important character—the glabrous stem—which could be incorporated in the possible hybrid produced from the interspecific cross with kenaf.

Preliminary work on species crosses had been done by Toxopeus (1947) although he was not able to produce a hybrid.

Singh *et al.* (1938) also did some interspecific crosses between *Hibiscus ficulneus* L. and *Hibiscus esculentus* L. from the systematic standpoint of determining the affinity existing between different *Hibiscus* species.

This experiment was conducted from November, 1958, to January, 1960 on the lot adjacent to the department of Agricultural Botany. It has the following objectives: (1) to determine the possibility of crossing kenaf (*H. cannabinus* L.) with roselle (*H. sabdariffa* L.), (2) to incorporate in the possible hybrid produced the desirable characters of the two parental species, and (3) to evaluate the resultant hybrid from the standpoints of agronomic and morphogenetic characteristics of either parent.

MATERIALS AND METHODS

Kenaf, variety *vulgaris* [2] (fig. 1) was planted alternately with roselle, red variety [3] (fig. 2) in rows one foot apart. Differential planting was done to synchronize the date of flowering of both species. The kenaf seeds were sown on November 30, 1958 and the roselle seeds a week after. The young plants were thinned down to 20 to a row six inches apart when they were about 15 centimeters high. All the necessary agronomic cultural practices were followed.

Direct and reciprocal crosses were made when the plants were in bloom. The flowers were emasculated by a forcep in the afternoon a day before anthesis. All flowers from female and male parents were covered with glassine bags prior to emasculation and after pollination the next morning. The capsules were harvested 28-30 days after pollination. The seeds per capsule were counted for all fruit sets of the direct and reciprocal crosses.

The F_1 seeds were planted in rows one foot apart on May 16, 1959. The percentage of germination was noted. The hybrid plants were selfed. They were also backcrossed to both parents to obtain any possible seed set and intensify the desired characters.

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[2] Seeds were from the Economic Garden, Los Baños, Laguna.
[3] Seeds were from the Botanical Garden, Bogor, Indonesia.

Fig. 1.—Portion of plant habit of one of the parental species (*H. cannabinus*).

Fig. 2.—Portion of plant habit of one of the parental species (*H. sabdariffa*).

The heights of the F_1 plants and the parental species were measured when they were 130 days old. Morphological and agronomic characters were also observed. The pollen and stomatal size was measured under the low and high power objective of the microscope. The frequency of abortive and non-abortive pollen was noted.

Ten representative stem samples from each of the hybrid and parental species were retted in a tank full of water for the determination of the tensile strength. The breaking load of the fibers was determined with the Scott Tester IP-2Tensilgraph using samples of single-fiber strands. The tensile strength of the fibers expressed in breaking length was computed in accordance with the formula of Botkin, Shires and Smith (1950).

RESULTS AND DISCUSSION

Table 1 shows the result of direct crosses between *H. cannabinus* L. and *H. sabdariffa* L. Seeds were produced, but they were shrivelled and non-viable. Examination of the seeds revealed that there was a small undeveloped embryo and a hardly discernible endosperm. The abnormal formation of the endosperm might be a possible explanation for the non-viability of the embryo.

TABLE 1.—*Number of capsules and seeds per capsule from direct cross between H. cannabinus L. and H. sabdariffa L.*

DATE POLLINATED	NO. OF POLLINATION	NO. OF FRUIT SET	NO. HARVESTED	NO. OF SEEDS/CAPSULE
2–17–59	3	1	1	1
2–18–59	5	2	2	4
2–21–59	5	2	1	4
2–23–59	5	2	2	3
2–24–59	8	1	0	0
2–28–59	6	1	1	2
3– 1–59	10	2	2	2
3– 3–59	10	1	1	4
3– 4–59	6	0	0	0
3– 5–59	15	4	2	2
3– 7–59	8	2	0	0
3– 9–59	8	4	1	1
3–10–59	5	1	0	0
3–11–59	6	0	0	0
3–15–59	4	1	0	0
Total	104	24	13	24
Mean	6.9	1.6	.86	1.8

Reciprocal crosses, however, produced a high fruit set with corresponding normal viable seeds (table 2).

TABLE 2.—*Number of capsules and seeds per capsule in the reciprocal crosses between H. sabdariffa L. and H. cannabinus* L.

DATE POLLINATED	NO. OF POLLINATION	NO. OF FRUIT SET	NO. HARVESTED	NO. OF SEEDS/CAPSULE
2–17–59	5	5	5	6
2–18–59	5	5	5	11
2–21–59	4	4	4	7
2–23–59	4	4	4	8
2–24–59	8	8	8	7
2–26–59	10	10	10	8
2–28–59	10	10	10	6
3– 1–59	15	15	14	10
3– 3–59	7	7	5	11
3– 4–59	8	8	8	9
3– 6–59	15	15	12	8
3– 7–59	12	12	9	8
3– 8–59	7	7	7	6
3–12–59	5	0	0	0
3–14–59	8	5	5	9
Total	123	115	106	114
Mean	8.2	7.6	7.1	7.6

Similar results were found by Deshpande (1938) in the interspecific crosses between kenaf and roselle. Medvedeva (1936) explained these results on the basis of their chromosome number, kenaf having 2n = 36 and roselle, 2n = 72. However, Narasinga (1935) and Ford (1938) found that some races of kenaf were tetraploids.

Based on the chromosome counts reported by Medvedeva (1936), it seems that the basic number of this genus is 18, where kenaf, having 36 chromosomes, would represent a diploid (2x) form and roselle with 72 chromosomes would constitute a tetraploid (4x). In the present experiments, the investigators tried to combine two chromosomal types:

(ovule) kenaf (36) x (pollen) roselle (72)—low set
(ovule) roselle (72) x (pollen) kenaf (36)— normal set

The above results generally agree with the chromosomal combination whereby an ovule parent with a low number of chromosomes fertilized by a pollen with a higher number of chromosomes produced

a low percentage of zygote. Diploid (2x) pollinated with tetraploid (4x) resulted in a low percentage of settings. On the other hand, a combination of tetraploid (4x) as material parent and diploid (2x) as pollinator gave a better setting of zygotes, as the resulting zygote was a triploid. The latter condition has been fully verified by morphological criteria in this kind of cytotype. In general, hybrid combinations of two chromosomal types are governed by either genic or chromosomal sterility or incompatibility, depending upon the genomic constitutions of the parental types (fig. 3).

Fig. 3.—Longitudinal section of the capsule of the parental species and F_1 hybrid showing aborted seeds in F_1 capsules. (a) *H. sabdariffa*, (b) F_1 hybrid, (c) *H. cannabinus*.

Thompson (1930) in his crosses between 14- and 21-chromosome species of wheat found that F_1 grains are plump when the 21-chromosome species is used as the female parent, and shrivelled when used as the male parent. He concluded that the seven extra chromosomes contributed to the normal development of the endosperm.

Two of the outstanding morphogenetic characters of the F_1 plants are vigor and sterility (fig. 4). Vigor was manifested by a significant increase in plant height. Sterility among the F_1 hybrid plants accounted for the difficulty in screening for possible good strains. Neither selfing nor backcrossing yielded any seeds. This could be explained if it is assumed that the F_1 hybrids are triploids. Although no cytological investigation was done, a microscopical examination of the stomatal size, size of pollen grain and fertility of pollen was made. All these criteria, except stomatal size, tended to verify its triploid condition.

Fig. 4.—F_1 hybrid of *H. cannabinus* × *F. sabdariffa*.

DESCRIPTION OF THE HYBRID

Agronomic Characters

Percentage of germination. Like the female parent, the F_1 seeds had a very low percentage of germination. This could be due to the very small size of the seeds and the thick seed coat.

Susceptibility to pests and diseases. Harm caused by pests and diseases is negligible although kenaf was slightly attacked by the common cotton stainer.

Height. As table 3 indicates, there is a significant increase in height among the hybrid plants, becoming very much taller than either parent.

Tensile strength of the fiber. Table 4 shows that there is no significant difference in the tensile strength of fiber between the F_1 hybrid and the parental species. The mean tensile strength of the hybrid is intermediate between parental species.

Days to flowering. The F_1 plants flowered later than either parent.

TABLE 3.—*Height of parental species and hybrid at 130 days of age (in centimeters)*

	H. SABDARIFFA	HYBRID	H. CANNABINUS
1	218.0	251.3	232.4
2	216.4	256.4	244.2
3	227.5	234.5	236.8
4	218.6	247.1	234.6
5	221.7	240.3	229.6
6	223.2	244.6	231.5
7	231.5	237.2	241.2
8	224.6	242.0	237.4
9	226.0	245.1	235.7
10	214.1	249.7	228.0
Total	2221.6	2448.2	2350.9
Mean	222.16	244.82	235.09

S.V.	D.F.	SS	MS	COMP. F-VALUE	PROBABILITY	HSD .05	HSD .01
Among species	2	2584.44	1292.22	39.00	<.01	6.23	7.93
Within species	27	894.51	33.13				
Total	29	3478.96					

Morphogenetic Characters

Stem. Trichomes are numerous on the stem and petiole of the hybrid. Thus, prickly condition could be considered as a dominant character.

TABLE 4.—*Tensile strength in kilometers of parental species and hybrid fiber*

	H. SABDARIFFA	HYBRID	H. CANNABINUS
1	32.50	32.50	34.25
2	29.25	31.50	33.25
3	30.75	33.50	38.50
4	36.50	35.75	34.00
5	32.50	31.00	30.00
6	35.50	36.50	31.00
7	34.00	37.25	36.25
8	29.50	30.75	35.25
9	33.50	28.50	33.50
10	35.25	33.25	37.00
11	29.75	35.00	36.25
12	35.00	36.50	38.25
13	35.50	28.50	37.00
14	30.00	36.50	34.50
15	29.50	33.75	31.00
Total	488.50	500.75	520.00
Mean	32.53	33.38	34.63

S.V.	D.F.	SS	MS	COMP. F-VALUE	PROBABILITY
Among species	2	33.63	16.81	2.06	.05
Within species	42	343.12	8.17		
Total	44	376.74			

Lobation of leaves. Like the male parent, the leaves of the hybrid are palmately to deeply lobed.

Joyner and Pate (1956), in their studies on the inheritance of leaf shape in kenaf, found that deep lobing is completely dominant over shallow lobing and the inheritance is on a monofactorial basis.

Stomatal size. Table 5 shows that the stomatal size of the hybrid is significantly smaller than either parent.

Pollen size and fertility. No significant difference in pollen size between the hybrid and the parental species exists as table 6 shows. Table 7 shows that there is a high percentage of abortive pollen (about 16 per cent). No abortive pollen was observed among the parental species.

TABLE 5.—*Stomatal size in microns of parental species and hybrid*

TRIALS [a]	H. SABDARIFFA	HYBRID	H. CANNABINUS
1	28.71	29.37	30.36
2	29.70	29.04	28.05
3	28.38	30.03	29.04
4	30.82	28.71	30.03
5	29.30	27.72	29.70
6	30.36	28.71	31.35
7	29.37	27.39	30.36
8	31.02	27.72	29.37
9	29.70	28.31	28.71
10	29.70	26.73	29.37
Total	297.06	283.73	296.34
Mean	29.71	28.37	29.63

[a] One trial is the average of 10 measurements.

S.V.	D.F.	SS	MS	COMP. F-VALUE	PROBABILITY	HSD .05	HSD .01
Among species	2	11.25	5.62	64.99	$<.01$	1.02	4.57
Within species	27	23.37	.86				
Total	29	34.62					

Capsule. The hybrid capsule is intermediate between the two parents with regards to the presence of bristles. This is similar to what Thombre (1958) obtained in his interspecific hybridization between *Mucuna pruriens* which has short pods with bristles, and *Mucuna Cochinchinensis* which has long, smooth pods without any bristle. Both species are legume plants. The hybrid is intermediate between the two parents with regard to bristles on the pod.

Comparison of morphological and agronomic characters between the hybrid and the parental species is summarized in table 8.

The relationship between the parental species and the F_1 could be stated as follows:

A. New characters in the hybrid

1. increased height (heterotic)
2. sterility (genic or chromosomal)
3. delayed time of flowering

TABLE 6.—*Pollen size in microns of parental species and hybrid*

TRIALS [a]	H. SABDARIFFA	HYBRID	H. CANNABINUS
1	150.28	140.40	143.20
2	130.24	142.08	140.24
3	156.85	143.56	144.60
4	145.40	133.16	141.72
5	130.24	145.00	134.32
6	152.24	149.48	142.84
7	130.24	139.14	134.32
8	140.60	139.12	139.85
9	139.20	146.15	135.80
10	130.24	139.12	139.85
Total	1405.53	1417.21	1396.74
Mean	140.55	141.72	139.67

[a] One trial is the average of 10 measurements.

S.V.	D.F.	SS	MS	COMP. F-VALUE	PROBABILITY	HSD .05	HSD .01
Among species	2	21.09	10.54	.226	.05	—	—
Within species	27	1260.49	46.61				
Total	29	1281.58					

TABLE 7.—*Frequency of abortive and non-abortive pollen grains of the F_1 hybrid*

TRIALS [a]	ABORTIVE	NON-ABORTIVE
1	7.5	1.1
2	17.4	.4
3	18.4	.5
4	28.0	.2
5	13.0	.8
6	17.7	.9
7	4.5	2.8
8	11.4	2.9
9	16.3	0
10	23.6	.3
Total	157.80	9.9
Mean	15.78	.99

[a] One trial is the average of 10 counts.

B. Intermediate characters (partial or incomplete dominance)

1. stem, petal and leaf color
2. presence of bristles on the capsule
3. size of epicalyx

C. Characters similar to the male parent, *Hibiscus cannabinus* L. (dominant)

1. prickly stem
2. staminal column and stigma
3. leaf lobation

The rest of the characters are similar to the female parent, *Hibiscus sabdariffa* L.

TABLE 8.—*Comparison of the agronomic and morpho-genetic characters between the F_1 hybrid and the parental species*

	H. SABDARIFFA	HYBRID	H. CANNABINUS
Height	1–2m.	2–4m.	2–3m.
Percentage of germination	50%	60%	85%
Days to flowering	125 days	130 days	118 days
Stem color	reddish-purple	intermediate	greenish
Prickles	absent	present	present
Leaf petiole	without prickles	with prickles	with prickles
Stipules	1 pair present	1 pair present	1 pair present
Color	purplish	intermediate	greenish
Lobing	shallow-lobed	deeply lobed	deeply lobed
Gland	reddish	reddish	greenish
Flower petals	purplish with dark purple center	yellow and purple-veined, center intermediate	yellow with purplish center
Stamen	staminal column short; stigma and anthers compact	staminal column and stigma long; anthers sparse	staminal column and stigma long; anthers compact
Calyx	bristles sparse; gland reddish	bristles intermediate; reddish gland	bristles thick greenish gland
Epicalyx	broad and hooked	intermediate	linear and pointed
Capsule	ovoid and pointed; bristles sparse	ovoid and pointed; bristles intermediate	globose; bristles thick
Seed	small in size; rough coat	shrivelled	bigger in size smooth coat

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THE ORGANIC MATTER, CRUDE WAX, TOTAL NITROGEN, P_2O_5, AND CaO CONTENTS OF FILTER PRESS CAKES FROM TEN SUGAR CENTRALS IN THE PHILIPPINES [1]

La Xuan Vinh and Leopoldo J. Villanueva

High percentages of crude wax (petroleum-ether extract) were obtained from the samples of press cake from 10 sugar centrals in the Philippines. The solvent used in wax extraction, the air-drying process, and the preservation of the press cakes with formalin apparently did not affect the total nitrogen, P_2O_5, K_2O, and CaO contents of the samples. Only slight differences in these constituents were found among the individual samples (not due to treatment). These differences may be traced to differences in varieties of cane milled and liming practices in the different centrals. The results obtained indicate that filter press cake can be a good fertilizer and soil conditioner, a promising source of commercial wax, and probably a suitable supplement for poultry and livestock feeds, provided it is properly prepared.

Filter press cake is an undesirable by-product of cane sugar manufacture. It is the soft, spongy, dark brown-black material resulting from the flocculates, "bagacillo", sugars, cane wax, albuminoids, coagulated colloids, phosphate of lime, sand and soil in the juice, and other sediments in cane juice after clarification and filtration. Hence, the name "filter press cake", "mud cake", or "factory mud".

In 1958-1959, the sugar mills in the Islands produced 237,784.05 [3] metric tons of this by-product. The disposal of this bulk of filter press cake by sugar centrals has always been a serious problem. If this filter press cake is utilized, instead of being dumped and made to decompose and rot in the factory premises, rivers and creeks, huge savings to the industry will result, and a good relationship between the sugar central and the people in the surrounding areas,

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[1] Central Experiment Station Contribution No. 2053. Read in the 278th meeting of the Los Baños Biological Club, June 30, 1960, and in the Philippine Sugar Technologists Convention, August 25, 1960.

[2] The authors are grateful to the sugar centrals concerned for furnishing the filter press cakes used in this study, and to the 1959 graduates in Sugar Technology for collecting the samples.

[3] Production figures. Sugar News **35** (9): 426 (1959).

who have long been irritated by the smell of the waste material, will be established.

In searching for ways of utilizing filter press cake, the determination of its composition is an important requisite. The results of press cake analyses will provide the necessary information for its ultimate utilization.

Samuels and Landrau, Jr. (1955), after determining the fertilizing value of press cake on sugar cane, vegetable crops, root crops, and pineapple, claimed that it may be used advantageously as fertilizer, especially on soils requiring nitrogen and phosphorus.

Owen (1954) believes that dewaxed, re-treated filter press mud stimulates plant growth, and is an excellent plant food for bacteria. He stated that this mud-contained food, an easily decomposable organic matter, possesses a high potential factor for microflora.

Rosales (1954) pointed out the possibilities of filter press cake as a source of wax, and the dewaxed cake as an excellent material for "organiculture". He cited the rising demand of all kinds of waxes, especially vegetable, for industrial purposes in the United States and other countries. He likewise mentioned the establishment of sugar cane wax recovery plants as early as 1951 in Java and in South Africa; the exportation of sugar cane wax to London in 1917 (Howes 1936), and of 6,000 tons in 1924 by South Africa (Case, 1927), the employment of the Rhodes and Swenson (1941) process of extracting wax directly from freshly discharged filter press cake in Cuban factories. He asserted that about 40 per cent of the cane wax is extracted with the juice while about 60 per cent remains in the bagasse. In his experiments at Central Don Pedro, he obtained an average of 14.60 per cent crude wax on the air-dried filter press cake. He summarized the Swenson war refining process as used at Gramercy, Louisiana, U.S.A.

Locsin (1953) claimed that filter press cake is a valuable fertilizer for the phosphate-deficient red soils of the Victorias and Manapla districts of Negros Occidental. When applied at the rate of 75 to 100 tons of wet cake per hectare, it can replace fertilization with 500 kilograms of ammophos (16-20-0), with a residual effect of about half of this amount for the following crop. He found the cake to have 2.01 per cent nitrogen (elemental), 3.70 per cent P_2O_5, 0.28 per cent K_2O, and 3.22 per cent CaO calculated on oven-dry basis.

Hernandez-Medina *et al.* (1953), in their study of the beneficial effects of filter press cake on pineapple yield under field conditions, found that larger fruits were obtained from plots receiving bigger amounts of filter press cake than from plots receiving smaller amounts. When at least 32 tons of press cake were applied to the acre, there was no need to apply more than half of the fertilizer ordinarily used in the pineapple fields. Their work on the effect of filter press cake on soil properties showed that the continuous and steady application of large quantities of organic matter for a number of years has produced a better state of aggregation, consequently forming a larger proportion of macroaggregates capable of holding considerable quantities of water at relatively low tension.

Misra *et al.* (1950) found that press cake had a more favorable effect on the growth of azotobacter population that promoted nitrogen fixation than oil cake or manure.

Walawalker (1949) reported that burning limed press mud gave a lime content of low quality (37.1 to 74.5% CaO, oven-dry basis).

Bonilla (1948) reported that a pilot plant using toluene as solvent recovered wax amounting to five per cent of dry cake from a Puerto Rico sugar mill.

In India, Darwalla and Kamath (1946) obtained a patent to make mineral wool from press cake and sand.

Balch (1945), working on mud press cake from Louisiana canes, obtained as high as 11.67 per cent purified wax from certain varieties and as low as 3.15 per cent or less from others. In 1945, he obtained a patent for a process of extracting crude wax with benzene, and removing the fatty portion with acetone.

Riollano (1943) reported that a significant increase in yield of vegetable crops was obtained when 20 tons of filter press cake were applied to the furrows alone. This increase was found to be equivalent to the yield obtained with complete fertilizer having 200 pounds NH_3, 200 pounds P_2O_5, and 200 pounds K_2O.

Aswath, Rao, and Gupta (1941), in extracting wax, used petroleum ether for initial extraction and purified the extract by treatment with nitric acid.

Guanzon (1927) analyzed press cake from the Canlubang Sugar Central and extracted wax, using alcohol and gasoline as solvents.

He claimed that "the residue from the extraction has some fertility value; the fertilizer constitutents of the cake being unaffected by the solvents in the process of extraction."

Deerr (1921), in his study of the fertilizing value of press cake, claimed that the material is rich in nitrogen and phosphate.

The objects of this work were (1) to determine the organic matter, crude wax, total nitrogen, P_2O_5, K_2O, and CaO contents of the fresh (formalin-preserved) and the air-dried filter press cakes from 10 sugar centrals in the Philippines, (2) to determine the total nitrogen, P_2O_5, K_2O, and CaO contents in the residues (dewaxed cakes) from wax extraction, and (3) to provide the necessary data for the ultimate utilization of filter press cake, i.e., as fertilizer and soil conditioner, as a source of commercial wax, and as a supplement, if properly prepared, to livestock and poultry feeds.

The work was done in the Department of Agricultural Chemistry from June, 1958, to October, 1959.

MATERIALS AND METHODS

Air-dried and fresh (formalin-preserved) samples were collected between the latter part of November, 1958, and early April, 1959, from 10 sugar centrals in the Philippines:

Centrals	*Location*
Central Azucarera de Tarlac	San Miguel, Tarlac
Pampanga Sugar Development Co.	San Fernando, Pampanga
Central Azucarera Don Pedro	Nasugbu, Batangas
Victorias Milling Company	Victorias, Negros Occidental
Hawaiian-Philippine Co.	Silay, Negros Occidental
Talisay-Silay Milling Co.	Talisay, Negros Occidental
Central Azucarera de la Carlota	La Carlota, Negros Occidental
Binalbagan-Isabela Sugar Co.	Binalbagan, Negros Occidental
Central Azucarera de Bais	Bais, Negros Occidental
Central Santos-Lopez Co.	Barotac Nuevo, Iloilo

All these centrals use clarification by defecation with lime and heat. Lime was used at the rate of 0.30 to 0.65 kilo of available CaO per ton of cane milled.[4]

[4] Weekly comparative run reports of all mills in the Philippines. Philippine Sugar Association Reports Nos. 1-40, 1958-1959 crop.

Preparation of sample. Fresh cakes were preserved in air-tight Mason jars using about 2 ml. of formalin per 500 gm. of sample. Samples collected from each central were subsampled until suitable sample sizes for analysis were obtained. For air-dried filter press cakes, the samples were ground until about 90 per cent of the particles passed through a 40-mesh sieve. The samples were then stored in air-tight Mason jars until analyzed.

Dewaxing of air-dried and fresh cakes. Portions of the air-dried and fresh (formalin-preserved) filter press cake samples from each of the 10 sugar centrals were oven-dried. Then 5 grams from each sample were dewaxed with petroleum ether in Soxhlet extractors for about eight hours or until the ether in contact with the cake was clear. The residue obtained after extraction was freed from ether in the water bath, then in the oven at 100°C. for 30 minutes. The dried materials, referred to in this paper as dewaxed cakes, were then wrapped in filter papers and stored in desiccators. The total nitrogen, P_2O_5, K_2O, and CaO contents of the dewaxed cakes were then determined, using the same methods, described below, for air-dried and for fresh (formalin-preserved) press cakes.

Removal of formalin in fresh (formalin-preserved) filter press cake. Fresh filter press cake was exposed to the atmosphere for 30 minutes to allow formalin to escape before weighing for analysis. The cake was then placed into a convenient air-tight bottle and weighed.

Moisture determination. Air-dried and fresh (formalin-preserved) filter press cakes were dried in the oven at 100° to 105°C., at atmospheric pressure until constant weight was reached. Since temperatures higher than 105°C. might decompose the sugars in the cake, an oven with a thermostat was used.

Organic matter determination. The modified method of Peech (1947) was used for convenience because of rapidity in determining the organic matter content of the cakes. A weighed quantity of cake containing about 50 mg. of organic carbon was transferred into a 500 ml. Erlenmeyer flask, and 20 ml. of N potassium dichromate was added. Rapidly, 20 ml. of concentrated sulfuric acid was added, directing the stream into the solution. The flask was immediately swirled by hand for one minute and then allowed to stand on a sheet of asbestos for about 30 minutes. Two hundred ml. of water, 10 ml. of phosphoric acid, and 0.5 ml. of barium diphenylaminosulfonate

indicator were added. Titration against ferrous sulfate was done until the solution was purple or blue, then ferrous sulfate was added in small amounts of about 0.5 ml. until the color flashed to green. The results were given as organic matter (Walkley and Black's values).

Petroleum-ether extract. Air-dried and fresh (formalin-preserved) cakes were first oven-dried. Then 0.2 gm. of the oven-dried cake was wrapped in filter paper and extracted with petroleum ether in a semi-micro Soxhlet extractor for eight hours. The fatty constituent was determined by difference.

Nitrogen determination. The modified Kjeldahl method prescribed by the A.O.A.C. was followed.

Ashing and preparation of extract solution for the determination of K_2O, P_2O_5, and CaO. Ashing was done according to the method prescribed by the A.O.A.C. (1955) for plant nutrients analysis.

Determination of P_2O_5 and CaO. The methods adopted by the A.O.A.C. for plant materials were used.

Determination of K_2O. Since H_2PtCl_6 was not available, the A.O.A.C. method was not adopted. Instead, the method given by Brown, Robinson, and Browning (1938) was followed.

To 1.5 ml. of 95 per cent ethyl alcohol in a 15-ml. centrifuge tube was added a 5-ml. aliquot (containing about 0.5 mg. K) of the potassium solution. Two ml. of sodium cobaltinitrite were then added dropwise with continuous shaking. The tube was allowed to stand for one hour at a temperature of 20° to 25°C. It was then centrifuged for about 10 minutes at about 2000 rpm so that the precipitate was firmly packed at the bottom of the tube. The supernatant liquid was poured off and the tube allowed to drain for about five minutes. The precipitate was washed with 5 ml. of 70 per cent alcohol, breaking up the bulk of the precipitate by forcing the wash solution, in a fine stream, from a pipette. The tube was again centrifuged for about five minutes and drained as before. The precipitate was then dried for 30 minutes at 80° to 85°C. to remove all the alcohol.

Then, to the precipitate in the tube, 6 ml. of the ceric sulfate reagent and 1 ml. of 1:1 sulfuric acid were added. The tube was heated in a water bath at 90° to 100°C. until all the precipitate was oxidized, as indicated by the disappearance of the precipitate (usually within 5 minutes). An excess of ceric sulfate was maintained

throughout the reaction. The tube was cooled to room temperature and the excess ceric sulfate was titrated against ferrous ammonium sulfate, using one drop of the *o*-phenanthroline ferrous complex as indicator. The end point was noted to be very sharp; the color of the solution changed from pale blue to red.

Standard solution of KCl was prepared and its aliquot was analyzed as check to determine the factor of conversion.

RESULTS AND DISCUSSION

Composition of the cake samples. The average percentage obtained from air-dried and fresh cakes, respectively, were: for moisture, 8.61 and 70.28 per cent; organic matter (Walkley and Black's values), 83.77 and 82.96 per cent; and crude wax (petroleum-ether extract), 11.34 and 11.22 per cent, all on oven-dry basis (table 1).

TABLE 1.—*The moisture, organic matter, and crude wax (petroleum-ether extract) contents of air-dried and fresh (formalin-preserved) filter press cakes from ten sugar centrals in the Philippines*

SAMPLE (AND ORIGIN)	MOISTURE		ORGANIC MATTER (WALKLEY AND BLACK'S VALUES)		CRUDE WAX (PETROLEUM-ETHER EXTRACT)	
	Air-dried	Fresh (Formalin-preserved)	Air-dried	Fresh (Formalin-preserved)	Air-dried	Fresh (Formalin-preserved)
	per cent	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*
1 (Talisay, Silay)	10.45	70.33	85.28	78.53	13.80	12.80
2 (Hawaiian-Philippines)	8.20	79.77	85.99	83.54	12.55	13.00
3 (Victorias)	6.77	72.70	83.60	83.70	11.50	11.40
4 (Bais)	7.37	63.47	80.14	82.26	7.30	7.10
5 (PASUDECO)	9.46	63.57	86.61	84.68	11.50	11.10
6 (Don Pedro)	9.84	62.98	85.23	86.08	11.95	11.90
7 (Santos-Lopez)	7.95	77.78	85.25	85.91	11.00	11.00
8 (Tarlac)	7.38	72.33	84.01	82.36	11.10	11.50
9 (BISCOM)	8.58	68.96	77.81	79.44	12.35	12.40
10 (La Carlota)	10.12	70.91	83.76	83.05	10.35	10.00
Averages	8.61	70.28	83.77	82.96	11.34	11.22

The average percentages of the other constituents, on oven-dry basis, were: for total nitrogen, 1.14 and 1.10; phosphoric acid, 3.58 and 3.67; potash, 0.44 and 0.42; and lime, 4.71 and 4.50 per cent for air-dried and fresh (formalin-preserved) cake samples, respectively (tables 2, 3, and 4). The dewaxed air-dried and fresh (formalin-preserved) samples, on oven-dry basis, gave respectively, 1.05 and 1.06

TABLE 2.—*The total nitrogen and phosphoric acid (P_2O_5) contents, on oven-dry basis, of air-dried (oven-dried and dewaxed), and fresh (formalin-preserved, oven-dried and dewaxed) filter press cakes*

SAMPLE (AND ORIGIN)	TOTAL NITROGEN				PHOSPHORIC ACID (P_2O_5)			
	Air-dried		Fresh (formalin-preserved)		Air-dried		Fresh (formalin-preserved)	
	Oven-dried	Dewaxed	Oven-dried	Dewaxed	Oven-dried	Dewaxed	Oven-dried	Dewaxed
	per cent	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*
1 (Talisay-Silay)	0.94	0.67	0.85	0.70	2.39	1.97	2.79	2.65
2 (Hawaiian-Phil.)	1.35	1.20	1.09	1.22	4.54	3.79	4.07	3.87
3 (Victorias)	1.45	1.26	1.17	1.25	3.37	3.28	3.09	3.25
4 (Bais)	0.58	0.50	0.60	0.50	2.39	2.32	2.31	2.37
5 (PASUDECO)	1.21	1.11	1.14	1.07	5.33	4.60	5.24	5.01
6 (Don Pedro)	1.14	1.19	1.29	1.21	3.99	4.65	4.17	4.79
7 (Santos-Lopez)	1.18	1.26	1.23	1.24	3.21	3.06	3.82	2.96
8 (Tarlac)	1.10	1.19	1.29	1.17	4.16	4.01	4.87	4.76
9 (BISCOM)	1.16	1.23	1.23	1.23	4.06	4.00	3.78	4.00
10 (La Carlota)	1.31	0.89	1.10	0.98	2.36	2.42	2.57	2.05
Averages	1.14	1.05	1.10	1.06	3.58	3.41	3.67	3.57

TABLE 3.—*The potash (K_2O) and lime (CaO) contents, on oven-dry basis, of air-dried (oven-dried and dewaxed), and fresh (formalin-preserved, oven-dried and dewaxed) filter press cakes*

SAMPLE (AND ORIGIN)	POTASH (K_2O)				LIME (CaO)			
	Air-dried		Fresh (formalin-preserved)		Air-dried		Fresh (formalin-preserved)	
	Oven-dried	Dewaxed	Oven-dried	Dewaxed	Oven-dried	Dewaxed	Oven-dried	Dewaxed
	per cent	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*
1 (Talisay-Silay)	0.22	0.20	0.18	0.22	3.05	2.98	2.71	2.87
2 (Hawaiian-Phil.)	0.23	0.22	0.24	0.21	4.22	4.13	4.23	4.16
3 (Victorias)	0.43	0.44	0.39	0.38	4.46	4.51	4.55	4.85
4 (Bais)	0.41	0.40	0.39	0.43	3.74	4.14	3.65	4.27
5 (PASUDECO)	0.42	0.39	0.41	0.40	5.19	5.12	4.56	5.27
6 (Don Pedro)	0.69	0.66	0.68	0.72	5.65	5.45	5.68	5.59
7 (Santos-Lopez)	0.60	0.63	0.58	0.59	4.49	4.29	4.27	3.65
8 (Tarlac)	0.52	0.52	0.51	0.54	6.11	5.49	4.78	4.07
9 (BISCOM)	0.45	0.41	0.39	0.39	5.40	5.02	5.21	4.82
10 (La Carlota)	0.38	0.39	0.42	0.38	4.79	4.58	5.34	4.29
Averages	0.44	0.43	0.42	0.43	4.71	4.57	4.50	4.38

TABLE 4.—*Average composition, on oven-dry basis, of air-dried, air-dried (dewaxed), fresh (formalin-preserved, and dewaxed) press-cake samples*

NATURE OF SAMPLE	COMPONENT					
	Total N	P_2O_5	K_2O	CaO	Organic matter	Crude wax (Petroleum-ether extract)
	per cent					
Air-dried	1.14	3.58	0.44	4.71	83.77	11.34
Air-dried (dewaxed)	1.05	3.41	0.43	4.57	(72.43)[a]	—
Fresh (formalin-preserved)	1.10	3.67	0.42	4.50	82.96	11.22
Fresh (dewaxed)	1.06	3.57	0.43	4.38	(71.74)[a]	—

[a] Minus crude wax.

per cent total nitrogen; 3.41 and 3.57 per cent P_2O_5; 0.43 and 0.43 per cent K_2O; and 4.57 and 4.38 per cent CaO (tables 2, 3, and 4). The organic matter of the dewaxed materials (obtained by subtracting crude wax from total organic matter) were 72.43 and 71.74 per cent for air-dried and fresh (formalin preserved), respectively (tables 1 and 4).

Comparison of the different cakes and residues from wax extraction. There were no significant differences in the oven-dry total nitrogen, P_2O_5, K_2O, and CaO contents of air-dried and fresh (formalin-preserved) cakes; and dewaxed air-dried and fresh (formalin-preserved cakes at $\alpha = .05$ level, which was selected because of the great variability of press cakes due to the heterogeneity of cane varieties milled in a central. The air-drying, preserving of the fresh samples with formalin, and extraction of wax with petroleum ether apparently did not affect the original total nitrogen, P_2O_5, K_2O, and CaO contents of the cakes (table 4).

Possible uses. The relatively high percentages of organic matter, total nitrogen, crude wax, phosphoric acid, potash, and lime in press cake indicate that it can be used as a fertilizer and soil conditioner, a feed supplement for livestock and poultry, and a good source of commercial wax. The organic matter in press cake can help improve soil texture and water-holding capacity.

Press cake, though lower in total nitrogen content than copra cake (4 per cent as reported by Prichett, 1924), is better than the latter since it also supplies a lot of P_2O_5, K_2O, and CaO.

Other observations. Slight variations in the percentages of each constituent in the samples, more particularly in the case of phosphoric acid, potash, and lime, might have been due to differences in soil conditions, milling and liming practices, and the heterogeneity of cane varieties milled.

SUMMARY AND CONCLUSIONS

1. No significant differences in the total nitrogen, P_2O_5, K_2O and CaO contents were found in the fresh, air-dried, and dewaxed cakes used in this study.

2. The fertilizer constituents of the cake appeared to be unaffected by the solvent used in the process of crude wax extraction, and by the formalin used in the preservation of the sample.

3. The high percentages of organic matter, total nitrogen, P_2O_5, and CaO of press cake suggest that it can be utilized as a fertilizer, soil conditioner and probably as a supplement for livestock and poultry feeds, provided it is properly prepared.

4. The high percentage of wax (petroleum-ether extract) in press cake suggests its commercial possibility as a good source of medium-hard commercial wax.

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A CORRELATION STUDY OF EGG SIZE TO FERTILITY, HATCHABILITY, AND CHICK SIZE; OF CHICK SIZE TO GROWTH AND MORTALITY; AND OF BODY SIZE OF HENS TO EGG PRODUCTION [1]

C. R. Arboleda, G. O. Ranit, and A. D. Yñiguez

WITH TWO TEXT FIGURES

Fertility and hatchability of Single Comb White Leghorn eggs were not affected by size. Egg size, however, was a determining factor in the size of the chicks hatched. The size of day-old chicks was not correlated with rate of growth. Mortality of the chicks within 12 weeks was not associated with the initial size of the chicks.

Single Comb White Leghorn pullet layers of varying sizes also laid eggs of varying number and sizes. Insignificant correlation was observed between the size of the hens and the number of eggs produced; however, the size of the eggs laid was statistically associated with the size of the hens.

Poultry raising on a commercial scale has brought about many problems, some of which are the determination of the relation of egg size to fertility, hatchability and chick size; chick size to rate of growth and mortality; and body size of hens to egg production. Single Comb White Leghorn chickens (S.C.W.L.) in the U.P. College of Agriculture are generally small but the size of eggs laid varies considerably. The usual practice is to incubate only those eggs that are almost uniform in size in order to produce uniform-sized chicks.

For commercial egg production the S.C.W.L. breed is commonly used in the country. Since the eggs laid vary considerably in size, this condition affects fertility, hatchability and size of chicks hatched. Enthusiastic beginners are confused on the size of chick to raise in order to get a favorable rate of body growth.

The relationship between body size of hens and egg production in the S.C.W.L. in the Philippines has not yet been sufficiently established. Campos (1955) reported a 300-egg production from S.C.W.L. pullets with an average weight of 1.42 kilograms. This

[1] Central Experiment Station Contribution No. 2054.

weight is far below the standard 2.04 kilograms for this particular breed (American Poultry Association, Inc., 1956).

At present, the Filipino consumers are not yet choosy about the size of eggs they buy from the market because of the limited supply of eggs and the high demand for them. With the growing interest of the people in raising poultry and the increase in poultry production, egg production will soon catch up with the demand and may eventually oversupply the market. Consumers would probably prefer big eggs, making it necessary for egg producers to adjust to such demands. Without sacrificing the quantity in egg production, how can big eggs be produced?

This study was conducted in the Poultry Husbandry Division, Department of Animal Husbandry, from August 15, 1959 to February 28, 1960, to determine the relation of size of eggs to fertility, hatchability, and chick size; size of chick to growth and mortality; and body size of hens to egg production.

MATERIALS AND METHODS

Eggs of the S.C.W.L. were used to correlate the size of eggs to fertility, hatchability and chick size. These chicks were raised and correlations between their day-old size to the subsequent sizes within intervals of two weeks for three months were derived. Mortality of the chicks was also correlated to the initial size of the chicks. Pullets of S.C.W.L. were used to correlate the body size with egg production.

Two methods of expressing the joint association were employed. The association between quantitative attributes such as size of eggs and chick size, chick size and growth, and size of hen and egg production was expressed in terms of the product-moment correlation coefficient.

The association between qualitative and quantitative attributes like size of egg to fertility, size of fertile eggs to hatchability, and size of chick to mortality was determined by the coefficient of contingency.

Correlation of size of eggs to fertility, hatchability and chick size. Eggs were collected from five pens of 15 layers and one rooster each. Eggs collected within a week were weighed individually on the Ohaus trip balance, and the weights were written in pencil

on the shell. Eggs below 44 and above 65.5 grams were excluded because they rarely appeared. Extra small eggs are usually yokeless and extra large eggs are frequently double-yoked. Normal eggs were then set in the Jamesway electric incubator. The incubating temperature was left constant at 99°F. After seven days of incubation the eggs were tested for fertility and viability through the electric tester. Weights of all infertile and D_1 (dead embryos at seven days) eggs were recorded. On the 14th day of incubation the eggs were again tested for viability and the D_2's were also recorded accordingly.

On the 18th day of incubation, the eggs were transferred from the incubating tray to the small wire crates (6 inches wide, 4 inches high and 24 inches long). Eggs of the same weight were placed in the same compartment of the 4-compartment wire crate. On the 22nd day, the chicks were taken out of the wire crates and weighed individually. The weights of the chicks were recorded according to the weights of the eggs from which they hatched.

Incubation was done in three replications at an interval of one week between replications. In the first replication only eggs from the five pens were collected, but because very few eggs were laid within a week by the hens from these pens, eggs from other pens were included in the experiment. The ratio between the male and female was 1:15 in all pens. In the second week of the first incubation, there was an intermittent electrical disturbance, causing a disruption of current in the incubator. This caused a high percentage of mortality in the fertile eggs. In the second and third replications however, incubation was undisturbed.

Correlation of chick size to rate of growth and mortality. Plastic wing bands (1/16 inch thick, 1/4 inch wide and 1 inch long) were pinned on the left wing of the chicks which were then weighed. The weights were recorded corresponding to their number on the wing bands.

The chicks were raised in the battery brooding cage heated by a 50-watt electric bulb. Floor space was provided at the rate of 0.25 square foot per chick.

During the first week of brooding, the chicks were given water and a little amount of feed. In the succeeding period, the chicks ate and drank as they pleased. The chicks were weighed forthnightly until the fourth week on the Ohaus trip balance.

After four weeks the chicks were transferred to the battery rearing cages provided with a sun porch. A chick occupied 1.2 square feet floor space. Feed and water were made available to the chicks at all times. A detecto balance was used in weighing them. In the twelfth week the chicks were weighed and the wing bands were removed.

During the brooding and rearing periods, some chicks died. A record of this was made according to the wing band identification of the chicks. The wing bands of a large number of chicks were removed. These were not considered in the samples although they remained in the cages.

Correlation of size of hens to egg production. Thirty-five pullet layers of the S.C.W.L. breed were picked from a flock having the same age and weighed individually on a detecto balance. Later, these layers were confined in the battery type laying cages. Feed and water were given at all times.

The number of eggs produced by each layer was recorded. After every collection, the eggs were weighed individually on the Ohaus balance.

The experiment was conducted from January 2 to February 28, 1960.

Housing and layer limitations brought about by the construction of the Poultry Division buildings of the college at this period necessitated the shortening of the observation period and the collection of samples.

RESULTS AND DISCUSSION

Fertility. The observed frequency of fertile and infertile eggs in the three trials is shown in table 1. In the first trial no egg was observed to be heavier than 60.5 grams. This is probably because only 98 eggs were observed. In the second and third trials, however, more eggs were included and all the sizes were represented. The occurrence of fertility did not follow a specific pattern with the varying sizes of eggs.

There was no relationship between size of eggs and fertility. The proportion between small and large fertile eggs was statistically the same. Even if there was an amount of contingency between the sizes of eggs and fertility the association was significant.

TABLE 1.—*Percentage of egg fertility, by different sizes*

TRIAL	FERTILITY	EGG SIZE (gm.)							
		45.0-48.0	48.5-50.5	51.0-53.0	53.5-55.5	56.0-58.0	58.5-60.5	61.0-63.0	63.5-65.5
I	Number fertile	12	32	19	20	7	2	—	—
	Percentage	80.00	94.11	100.00	95.23	100.00	100.00	—	—
II	Number fertile	8	40	36	35	33	15	10	2
	Percentage	72.73	100.00	90.00	89.74	91.66	93.75	90.90	0
III	Number fertile	7	34	41	47	39	20	12	3
	Percentage	100.00	91.89	89.13	93.87	100.00	96.24	85.71	100.00

The Chi-square analysis in table 1a shows that the computed Chi-square in all the trials did not exceed the tabular value at five per cent probability level.

TABLE 1a.—*Chi-square analysis and coefficient of contingency*

TRIAL	SAMPLE SIZE	FERTILE	INFERTILE	CHI-SQUARE (Xi^2)	DEGREES OF FREEDOM	COEFFICIENT OF CONTINGENCY
I	98	92	6	7.18 [a]	5	0.26
II	195	179	16	13.02 [a]	7	0.25
III	216	202	14	5.77 [a]	7	0.16

[a] Insignificant.

Hatchability. Table 2 shows the observed hatchability of fertile eggs. In all trials the proportion of the eggs that hatched to the dead fluctuated, instead of following a consistently increasing or decreasing trend. As long as the fertile eggs were normal, the hatchability was independent of the size. A comparison of the computed Chi-square value and the tabular value failed to show significance at five cent probability level.

Chick size. The size of chicks was highly dependent on, and directly proportional to the size of eggs from which the chicks hatched. (fig. 1a, lb, and 1c).

Even at the one per cent probability level, the computed correlation coefficients 0.436, 0.748 and 0.420 for trials I, II and III, respectively, still exceeded the tabular value.

Many workers in the United States have studied the effect of size to hatchability and have found conflicting results. Jull, Haynes and Warren as cited by Warren (Taylor, ed., 1949), each showed that small eggs hatched better than medium-sized eggs. Dunn (1922) contended that the hatchability of large eggs of both hens and pullets was below the average. On the other hand, Cañizares (1941) stated that there was no difference in fertility and hatchability of eggs of different sizes in the Los Baños Cantones chicken. According to Warren (Taylor, ed., 1949), these differences in results are due to the fact that all workers did not use the same standard for large egg size.

Correlation of chick size to growth and mortality. Table 3 shows the different correlation coefficients between size of chick and the growth at two weeks' interval for three months.

TABLE 2.—*Per cent hatchability of fertile eggs by different sizes*

TRIAL	HATCHABILITY	EGG SIZE (gm.)							
		46.0-48.0	48.5-50.5	51.0-53.0	53.5-55.5	56.0-58.0	58.5-60.5	61.0-63.0	63.5-65.5
I	Number hatched	8	20	17	13	4	0	—	—
	Percentage	66.66	62.50	89.47	65.00	57.14	0	—	—
II	Number hatched	7	35	29	31	32	13	8	0
	Percentage	86.75	87.50	80.55	86.50	96.96	86.66	80.00	0
III	Number hatched	6	31	35	41	35	16	9	3
	Percentage	85.71	91.17	85.36	89.13	89.74	80.00	25.00	100.00

TABLE 2a.—*Chi-square analysis and coefficient of contingency*

TRIAL	SAMPLE SIZE	LIVING	DEAD	CHI-SQUARE (Xi^2)	DEGREES OF FREEDOM	COEFFICIENT OF CONTINGENCY
I	32	62	30	7.78 [a]	5	0.38
II	178	155	23	8.54 [a]	7	0.21
III	202	176	26	3.68 [a]	7	0.13

[a] Insignificant.

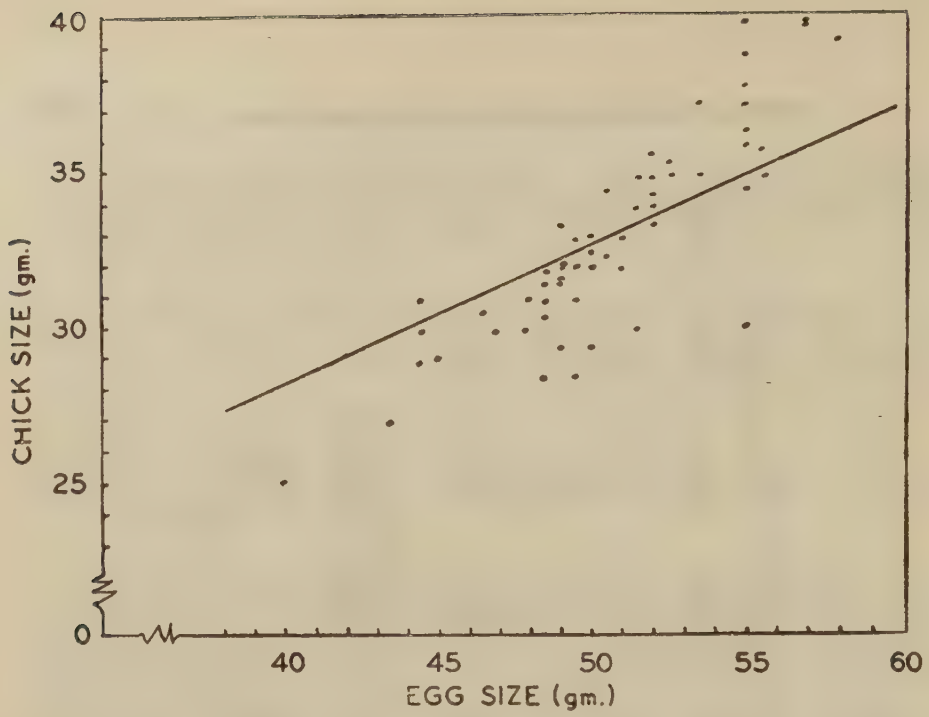

Fig. 1a.—Scatter diagram of the correlation of egg size to chick size in trial I ($r = 436$; $n = 63$).

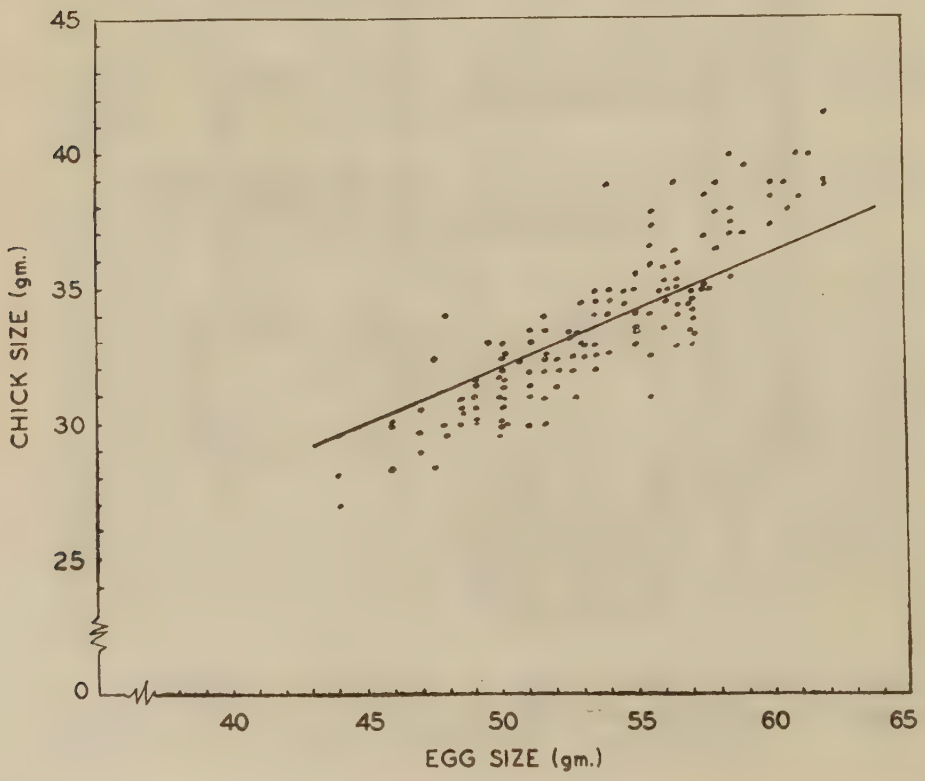

Fig. 1b.—Scatter diagram of the correlation of egg size to chick size in trial II ($r = 0.420$; $n = 170$).

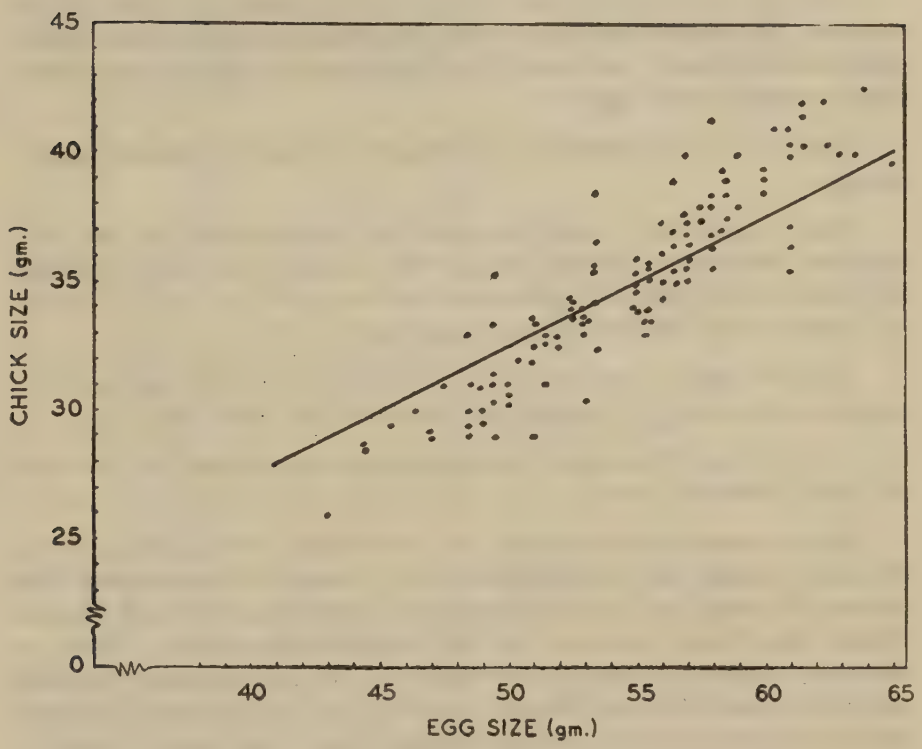

Fig. 1c.—Scatter diagram of the correlation of egg size to chick size in trial III ($r = 0.748$; $n = 152$).

TABLE 3.—*Correlation coefficient between size of chicks to its growth within three months at two weeks' interval*

TRIAL	TOTAL	MALE	FEMALE	AGE (WEEK)					
				2	4	6	8	10	12
I	34	17	17	0.164	0.095	0.038	0.138	0.082	0.094
II	35	23	12	0.171	–0.176	–0.102	–0.293	–0.131	–0.141
III	38	21	17	0.503	0.393	0.289	0.393	0.289	0.373

The general trend of the correlation coefficients was to decrease with the age of the chick except for trial I wherein the correlation coefficient followed an irregular increase and decrease from the 8th to the 12th week. Some chicks in the second trial whose initial weights were below the average grew even bigger than those chicks whose initial sizes were relatively large. This is shown by the negative correlation coefficients in the 4th to the 12th week.

The rate of growth of the chicks did not depend on their initial weights. The chicks would not grow big simply because their initial weights were heavy, nor would they grow poorly just because their initial weights were light.

Not one of the correlation coefficients in table 3 attained significance.

More chicks died in the first week of brooding and the last few weeks of rearing than in any other week. Table 4 shows the number of living chicks after 12 weeks and the mortality within 12 weeks as related to the different initial weights.

The life span of the chicks did not depend on their initial sizes. Large chicks, like the medium and small chicks, also died.

None of the Chi-square values attained significance at the five per cent probability level.

Many workers have studied the correlation of the early growth of chicks to egg size. Cañizares (1941) found that egg size of Los Baños Cantones did not influence the rate of growth of chicks. Others have found that egg size had a measurable positive effect on the early growth of chicks, but that this effect decreased rapidly after two weeks of age, thereby showing no appreciable influence on body weight at broiler age (Godfrey *et al.*, 1943; Kosin, 1952).

The first phase of this experiment showed that egg size exerted a very significant effect on chick size, but as the chick grew older, the initial chick size had little effect on the weight of the chick.

Correlation of size of hens to egg production. The weight of the hens converged closely to their average weight, although eggs became bigger as the time of observation lengthened. The average weight of the layers was 1.56 kilograms. Figure 2 shows the average number of eggs produced by different body sizes of hens, and its respective correlation coefficient.

The number of eggs produced by the hens of different sizes was so scattered that it did not follow any pattern. The size of eggs, however, was directly proportional to the size of hens. Large hens generally laid large eggs.

Other similar studies have shown varying results. Scott, as cited by Warren (Taylor, ed., 1949), observed a positive relationship between body size and weight of the yolk, and said that the size of

TABLE 4.—*Percentage of mortality of chicks by different initial sizes*

TRIAL	MORTALITY	INITIAL SIZE (gm.)						
		27.0-29.0	29.5-31.5	32.0-34.0	34.5-36.5	37.0-39.0	39.5-41.5	42.0-44.0
I	Number dead	3	0	4	3	3	—	—
	Percentage	41.42	0	22.22	41.42	41.42	—	—
II	Number dead	5	12	12	10	4	3	1
	Percentage	62.50	42.85	58.62	41.67	28.57	75.00	50.00
III	Number dead	0	6	11	12	5	2	1
	Percentage	0	54.54	39.28	35.29	27.77	18.18	38.34

TABLE 4a.—*Chi-square analysis and coefficient of contingency*

TRIAL	SAMPLE SIZE	LIVING	DEAD	CHI-SQUARE (X^2)	DEGREES OF FREEDOM	COEFFICIENT OF CONTINGENCY
I	48	35	12	4.29 [a]	4	0.29
II	109	57	52	4.54 [a]	6	0.20
III	108	71	37	5.66 [a]	6	0.22

[a] Insignificant.

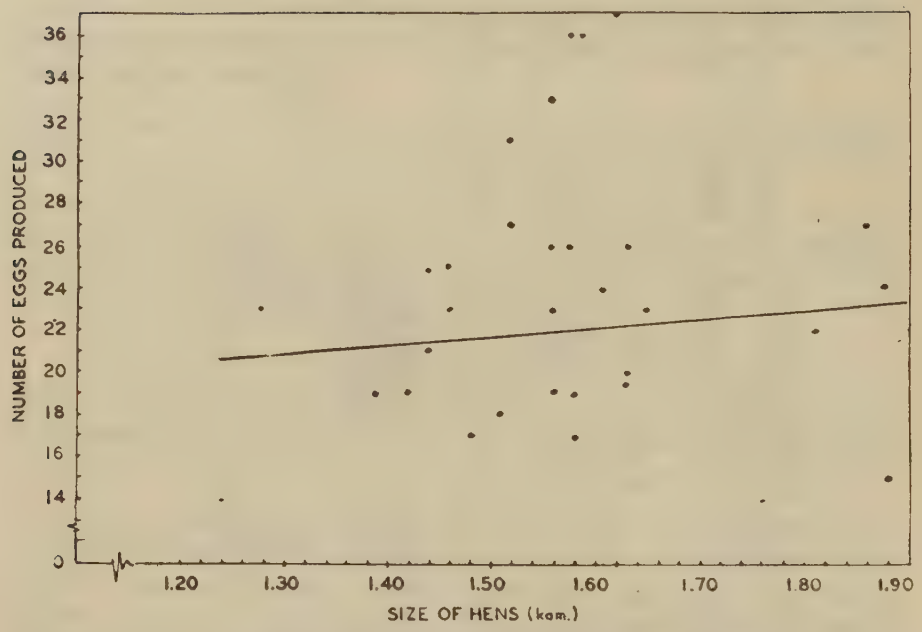

Fig. 2a—Scatter diagram of the correlation of body size of hens to number of eggs produced ($r = 0.103$; $n = 35$).

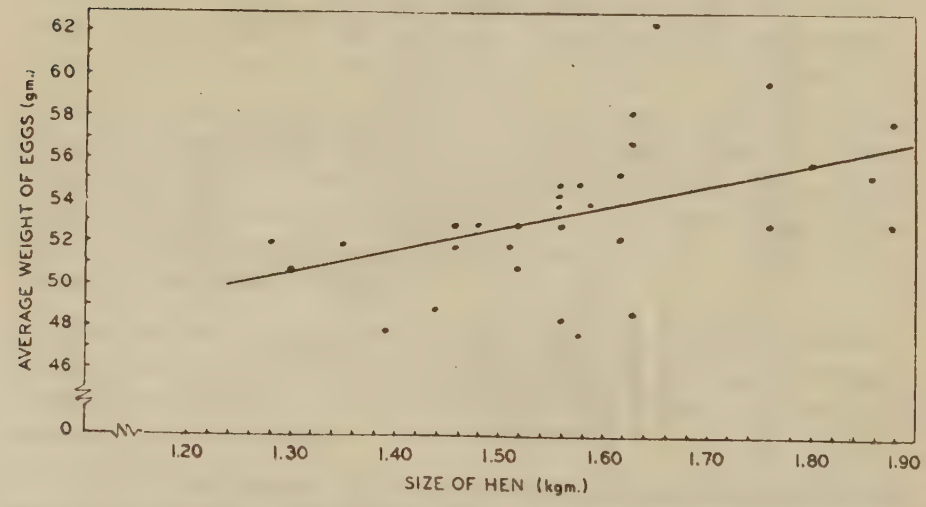

Fig. 2b.—Scatter diagram of the correlation of the body size of hens to the average weight of eggs produced ($r = 0.457$; $n =$ CE).

the yolk has long been known to be an important factor in determining egg weight. Campos (1955) stated that there is no direct relationship between body weight of each layer and average weight of egg produced, and the number of eggs laid by a pullet is often inversely correlated to body size. Blyth (1952) explained the inconsistency of the relationship of the number of eggs produced by a hen to the average weight. He stated that when hens laid eggs above the average number usually produced, the size of the egg was inversely correlated to the number, but this relationship failed to be true when the number of eggs laid was below the average.

SUMMARY

1. The degrees of association between fertility and size of normal eggs as measured by the contingency coefficient C were 0.26, 0.25, 0.16 in the first, second, and third trials, respectively. These degrees of association are insignificant.

2. Associations between hatchability and size of fertile eggs in trials I, II, and III were 0.28, 0.21, and 0.13, respectively. These degrees of association are also insignificant.

3. Chick size was found to be highly correlated with the size of eggs from which the chicks hatched. Correlation coefficients of 0.430, 0.420, and 0.784 were observed in trials I, II, and III, respectively.

4. The growth of chicks to 12 weeks was independent of the initial size of the chicks. Correlation coefficients of 0.094, –0.141, and 0.373 were observed for trials I, II, and III, respectively. All are insignificant.

5. Body size of hens was not a determining factor in the number of eggs produced. A correlation value of 0.103 was observed.

6. The size of eggs produced varied directly with the size of hens. A correlation coefficient of 0.457 was obtained.

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A SURVEY OF THE USE OF CHROMIC OXIDE AS INDICATOR FOR MEASUREMENT OF DRY MATTER INTAKE OF GRAZING CATTLE IN THE PHILIPPINES [1]

MAXIMO C. GACULA, JR., AMELIA B. LOPEZ,
AND LITO L. CALO

WITH TWO TEXT FIGURES

Chromic oxide recoveries of composited samples for both trials did not deviate significantly from 100 per cent. There was no significant difference between the measured dry matter intake and that estimated from the fecal chromic oxide analysis.

Chromic oxide concentration in the fecal samples obtained at different times of the day showed marked variability. However, the trend of the chromic oxide elimination pattern with respect to the time of the day indicated that accurate intake estimates could be derived from "grab" samples of feces taken at 6 a.m. and 4 p.m. Fecal samples procured during these hours and composited on an equal weight basis for a four-day collection period resulted in a reliable estimate of the dry matter intake for both hand-fed and grazing animals.

To appraise the feeding and economic value of forages, one must know the animal's rate of consumption of dry matter. However, under pasture or range conditions where animals are allowed to graze freely, no direct way of determining the amount of herbage consumed is known. Investigators have proposed the use of indirect methods, the underlying principle of which is that when digestibility of the herbage is known, the amount of feces voided can be used to calculate the amount of herbage consumed by the animal.

Fecal output may be obtained by the use of collection bags and harnesses, but these equipments affect the grazing behavior of the animal and have been reported to be laborious, time-consuming and not readily adapted for use with cows. Substituted for this practice was the use of an undigestible reference substance which, when fed to the animal in known quantities, may be employed to calculate the amount of feces voided per unit of time.

Of the reference substances used, chromic oxide has been noted to be an adequate indicator of the fecal output of grazing animals (Coup, 1950; Lancaster *et al.*, 1953; Kane *et al.*, 1953; Hardison and

[1] Central Experiment Station Contribution No. 2055. Project No. 1321.

Reid, 1955; and Smith and Reid, 1955). However, these workers conducted their studies under temperate conditions. The present study will test the validity of chromic oxide and consequently, the effectivity of this technique under Philippine conditions.

This study was conducted from October, 1958 to April, 1959 in the Department of Animal Husbandry, U.P. College of Agriculture. It aims 1) to determine the extent of recovery of chromic oxide in the feces, 2) to observe the chromic oxide elimination pattern of grazing cattle under local conditions, and 3) to check the accuracy with which the amount of dry matter intake can be determined using chromic oxide as an indicator.

MATERIALS AND METHODS

Trial I

Six steers with ages ranging from five to seven years were hand-fed with clipped forage (Para grass, *Brachiaria mutica*) twice a day, at approximately 6 a.b. and 5 p.m. Ten grams of chromic oxide were administered in gelatin capsules by the use of a balling gun at 4:30 p.m. each day of the 19-day experimental period. Records of the forage dry matter intake and fecal output during the seven-day collection period were kept. Three hundred grams of fresh fecal samples were secured every morning and evening for the analysis of chromic oxide. The sum of the morning and evening fecal collections was considered the total collection for the day. Composite samples for the entire collection period were also obtained for chromic oxide analysis.

Trial II

Two groups of three steers each were used. One group was hand-fed with clipped forage twice a day at 6 a.m. and 5 p.m.; the other group was allowed to graze on Alabang X (*Dichantium aristatum*) pasture, which was adjacent to the lot containing forage for hand-feeding. The experimental period included a preliminary period of 10 days and a collection period of four days. Ten grams of chromic oxide placed in a gelatin capsule were administered beginning the first day of the experimental period.

Grazing steers were fitted with collection bags (fig. 1) to facilitate total collection of feces. During the collection period, four-day composite samples and "grab" samples obtained at 6 a.m. and 4 p.m. were kept. On the second and last day of the collection period, fecal

Fig. 1.—An experimental animal with the attached collection bag.

samples were procured rectally at two hours interval from both groups to study the chromic oxide elimination pattern. The fecal samples obtained in trial I and trial II were analyzed in the Animal Nutrition Laboratory, Department of Animal Husbandry.

Method of analysis. To determine moisture and ash, three grams of sample were weighed in a porcelain crucible, dried in the oven to constant weight, ignited in the furnace at 700°C, cooled to room temperature in a dessicator and weighed.

To determine the chromic oxide level, a 0.15 gram ashed-sample was weighed in a nickel crucible with 0.25 gram sucrose and 0.50 gram KNO_3. About three grams of Na_2O_2 were added to the mixture. The contents were made to react over a very low flame. Complete fusion was attained by placing the nickel crucible in the furnace at 500°C.

After sufficient cooling, the crucible was placed on its side in a 600-ml. beaker containing enough distilled water to fully cover it. The beaker was covered with watch glass to prevent loss by spattering. When the melt was dissolved and the gas ceased to evolve, the crucible was rinsed with distilled water and the solution was filtered on fluted filtered paper into a 500-ml. iodine flask. The filtrate and washings were heated to gentle boiling for 30 minutes, then cooled

to room temperature (final volume about 250 ml.) After 25 ml. of 30 per cent H_2SO_4 and 0.5 gram KI were added, the flask was placed in the dark for five minutes and titrated with 0.1N $Na_2S_2O_3$ using starch as indicator.

Estimated herbage dry matter intake was derived from the following equations as presented by Smith and Reid (1955).

$$\text{a) Fecal output (gm.D.M./day)} = \frac{\text{Cr}_2\text{O}_3 \text{ consumed (gm./day)}}{\text{Cr}_2\text{O}_3 \text{ concentration of feces (gm./gm.D.M.)}}$$

$$\text{b) Herbage intake (gm.D.M./day)} = \frac{\text{Fecal output (gm.D.M./day)}}{\text{Per cent indigestibility}}$$

RESULTS AND DISCUSSION

Recovery of chromic oxide in feces. The recovery of chromic oxide in feces for trials I and II are summarized in table 1. Of the

TABLE 1.—*Percentage of chromic oxide recovery from composite feces voided during a four- to seven-day collection period*

		CHROMIC OXIDE		PER CENT RECOVERY [a]
		Intake (gm.)	Output (gm.)	
Trial 1	Hand-fed			
	1	10	10.24	102.4
	2	10	9.85	98.5
	3	10	10.65	106.5
	4	10	11.98	119.8
	5	10	10.38	103.8
	6	10	10.50	105.0
Trial 2	Hand-fed			
	1	10	9.68	96.8
	2	10	11.73	117.3
	3	10	9.00	90.0
	Grazing			
	4	10	9.98	99.8
	5	10	9.69	96.9
	6	—	—	— [b]
Mean recovery		10	10.33 ± 0.272	103.3 ± 2.727

Computed $t = 1.26$.
Probability $> .05$.

[a] Per cent recovery based on the 10 grams intake of chromic oxide and average fecal dry matter output per day.

[b] Animal number 6 had been counted out in the middle of the experiment due to a serious barbed wire injury, hence data were excluded.

10 grams of chromic oxide administered daily to the steers, the amount recovered in the feces ranged from 9.00 to 11.98 grams or a mean daily recovery of 10.33 ± 0.272 grams.

Smith and Reid (1955) reported the mean recovery of chromic oxide from totally collected feces composite samples as 97.5 ± 1.73 per cent. In this experiment, the mean recovery from the seven-day composite samples of totally collected feces was 103.3 ± 2.72 per cent, which apparently showed that chromic oxide passes the digestive tract unabsorbed and undigested, a characteristic greatly desired for indicators (Maynard, 1951). The per cent recovery in this experiment was found to be not significantly different from 100 per cent. It is therefore safe to assume that chromic oxide is completely indigestible and can be employed as indicator for fecal output determination.

Chromic oxide as indicator. The dry matter intake estimated from the chromic oxide concentration in the feces did not differ significantly from the measured intake (table 2). The mean difference was 0.123 kilogram. This seems to indicate that the intake computed from the analysis of chromic oxide in fecal samples voided during the 24-hour period and composited for seven days is a fairly reliable estimate of the actual dry matter intake.

TABLE 2.—*Comparison of measured and estimated dry matter intakes during a seven-day collection period*

ANIMAL NUMBER	DRY MATTER INTAKE (KG./DAY) FOR HAND-FED STEERS	
	Measured [a]	Estimated [b]
1	4.79	4.67
2	4.74	4.82
3	4.63	4.35
4	4.50	3.76
5	4.85	4.67
6	4.84	4.61
Mean	4.73 ± 0.137	4.48 ± 0.385

Computed $t = 2.09$.
Probability $> .05$.
[a] Actual intake.
[b] Intake based upon the chromic oxide concentration in the feces.

A comparison was made between the intake derived from the analysis of fecal samples voided during half-day periods and the measured intake (table 3). The intakes estimated from the half-day

TABLE 3.—*Comparison of intake derived from the analysis of fecal sampes voided during a half-day period with the measured intakes*

DATE	SAMPLING TIME	ESTIMATED DRY MATTER INTAKE FOR HAND-FED STEERS					
		1	2	3	4	5	6
2/13	Morning	4.89	8.60	6.50	3.56	5.51	6.84
	Evening	5.68	3.82	5.35	4.04	6.19	5.81
2/14	Morning	6.62	5.00	4.81	5.95	4.71	3.25
	Evening	9.00	6.45	6.73	8.93	4.53	3.51
2/15	Morning	4.75	4.02	5.53	4.20	4.75	4.52
	Evening	4.85	4.18	3.41	3.76	5.40	4.69
Average intake based upon analysis of half-daily samples of feces		5.96 ± 1.90	5.34 ± 1.86	5.27 ± 1.26	5.07 ± 1.27	5.18 ± 0.24	4.77 ± 1.36
Measured intakes		4.79	4.74	4.63	4.50	4.85	4.84

Computed $t = 3.49$.
Probability $< .05$.

samples of feces were significantly different from the measured intake. This corroborate the findings of Hardison and Reid (1953). The mean difference between the intakes was 0.653 kilogram.

Although the chromic oxide measured in feces representative of those voided during periods of seven days was found to offer an adequate index of the fecal voided, the application of this method of sampling necessarily requires a total collection of feces defecated. This mar the advantage of using chromic oxide in the estimation of intake under these sampling conditions. Hardison and Reid (1953) stated that if chromic oxide was excreted in the feces either uniformly or in a predictable manner, it would be possible to supplant the total collection of feces by a series of "grab" samples taken a few times in one or more days. This would eliminate the necessity for fecal collection bags, allow more use of female animals, and save time and labor in experimentation. Since diurnal variation of the chromic oxide concentration in feces has been observed (Kane *et al.*, 1952; Hardison and Reid, 1953; and Kameoka *et al.*, 1956) "grab" sampling can be effectively used only if the chromic oxide elimination pattern in feces is known. To verify this possibility under our conditions, trial II was conducted to determine the chromic oxide elimination pattern in feces of both hand-fed and grazing animals.

Chromic oxide elimination-time pattern in feces. In determining the chromic oxide elimination-time pattern, fecal samples obtained at two-hour intervals during the two collection days from both treatments were compounded and the analyses expressed in per cent recovery (fig. 2).

Hardison and Reid (1953) obtained a recovery of 80 to 130 per cent for hand-fed animals and 50 to 180 per cent for grazing animals. In this study, chromic oxide recovery in feces from "grab" samples taken at various times of the day varied from 66.66 per cent to 233.33 per cent for grazing animals, and 80.48 per cent to 112.19 per cent for hand-fed animals. The lowest recovery rate of fecal chromic oxide occurred at 6 a.m. and the highest at 2 p.m. for grazing animals; for the hand-fed group, the lowest was at 8 a.m. and the highest at 8 p.m. and 10 p.m.

A wider range of fecal chromic oxide variation from 100 per cent recovery was observed among the grazing animals than among the hand-fed animals. The distinct variation in these two patterns could be traced to the different intake levels between the two treatments and the time herbage was consumed.

The chromic oxide patterns formed in both groups were studied to select the time of the day during which one could collect "grab" fecal samples that would approximate 100 per cent recovery of the ingested chromic oxide.

Under grazing conditions, the sampling combinations close to 100 per cent recovery were 10 a.m. and 8 p.m. with an average recovery of 99.75 per cent; 6 a.m. and 4 p.m. with 103.33 per cent; 6 a.m. and 12 noon with 101.66 per cent, and 12 noon and 4 a.m. with 106.66 per cent. Among the hand-fed animals, 12 noon and 4 p.m. had a recovery of 102.44 per cent; 6 a.m. and 4 p.m. with 98.78 per cent; 6 a.m. and 8 p.m. with 99.99 per cent, and 10 a.m. and 8 p.m. with 97.56 per cent. Of the various sampling time combinations studied, "grab" sampling at 6 a.m. and 4 p.m., and 10 a.m. and 8 p.m. were found to have close to 100 per cent recovery in both hand-fed and grazing animals. However, in our type of livestock management, 6 a.m. and 4 p.m. seemed to be the most convenient times for "grab" sampling. Linkous *et al.* (1952) considered 6 to 8 a.m. and 6 to 8 p.m. collection, while Kane *et al.* (1952) recommended 1 p.m. to 3 p.m. as the best time for daily sampling. The present data conform favorably with the findings of Hardison and Reid (1953), and Smith and Reid (1955).

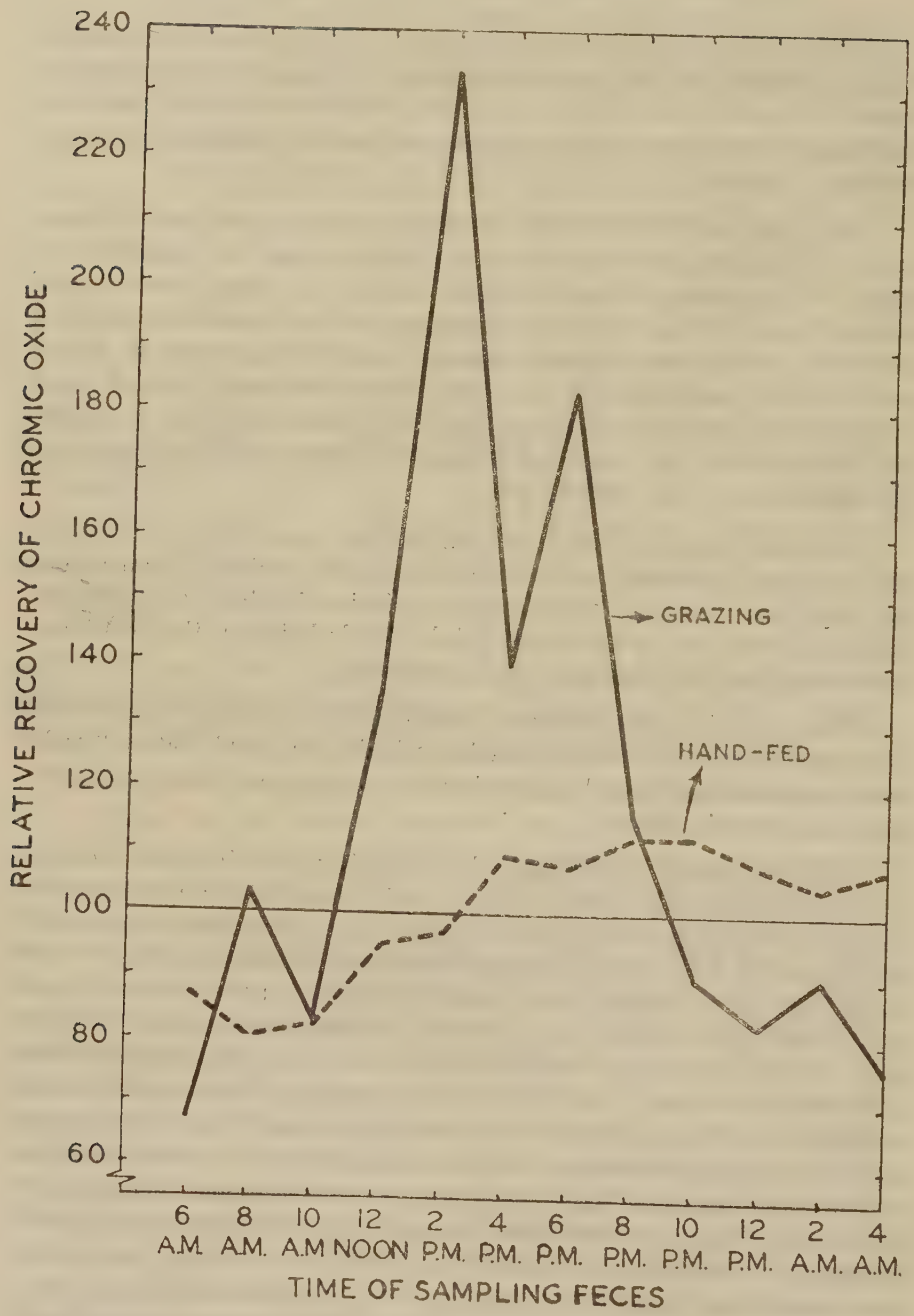

Fig. 2.—Chromic oxide elimination-time pattern for hand-fed and grazing steers administered chromic capsules at 4:30 p.m.

Grab sampling at 6 a.m. and 4 p.m. Chromic oxide recovery in the feces obtained from "grab" samples taken at 6 a.m. and 4 p.m. averaged nearly 100 per cent. For hand-fed animals, the chromic oxide recovery at 6 a.m. was 87.80 per cent and at 4 p.m., 109.75 per cent, or an average of 98.75 per cent; for grazing animals, 66.66 per cent and 140 per cent or an average of 103.33 per cent. Despite this variability in the chromic oxide concentration of the individual "grab" samples, the chromic oxide concentration of feces composited from collections at these two times were very similar to those of the total feces collected during the corresponding 24-hour period. Hardison and Reid (1953) reported an average recovery of 104 per cent in the hand-feeding trials and 99.95 per cent in the grazing trials. In a similar study, Smith and Reid (1955) reported a mean recovery rate of 100 ± 0.87 per cent of ingested chromic oxide from the combined feces sampled at 6 a.m. and 4 p.m.

In this experiment, a test for estimating the accuracy of herbage dry matter intake as a result of sampling feces at 6 a.m. and 4 p.m. was made. The dry matter intake derived from the "grab" samples and four-day composite samples and from the measured intake taken at the same period did not differ significantly (table 4). These were observed in both hand-fed and grazing animals. This suggests that compounding "grab" samples taken at 6 a.m. and 4 p.m. for four or more days would give an equally accurate estimate

TABLE 4.—*Comparison of intakes derived from the analysis of feces taken at 6 a.m. and 4 p.m. and a four-day period with the measured intakes*

DATE	SAMPLING TIME	DRY MATTER INTAKE (KG./DAY)					
		Hand-fed steers			Grazing steers		
		1	2	3	4	5	6
3/28	6 a.m.	4.82	7.30	9.97	9.15	9.21	—
	4 p.m.	5.90	7.77	8.60	7.13	9.59	—
3/29	6 a.m.	6.40	5.07	4.33	7.47	7.83	—
	4 p.m.	6.05	2.83	3.81	6.22	4.81	—
3/30	6 a.m.	6.34	7.20	9.11	9.23	9.52	—
	4 p.m.	6.98	7.39	8.65	8.38	9.93	—
3/31	6 a.m.	6.07	6.43	8.04	10.74	10.74	—
	4 p.m.	5.32	5.72	7.64	7.65	7.44	—
Mean		5.98	6.22	6.56	8.24	7.50	—
		± 0.234	± 0.169	± 0.791	± 0.504	± 0.794	—
Estimated intakes based on analysis of four-day composite sample		6.20	5.16	6.96	8.54	8.24	—
Measured intake		6.00	6.06	6.24	8.50	8.25	—

Analysis of variance

SOURCE OF VARIATION	DF	SUM OF SQUARES	MEAN SQUARE	COMPUTED F-VALUE	PROBABILITY
Intake determination methods	2	0.042	0.021	0.013	>.05
Animals within methods	12	18.790	1.565		
Total	14	18.832			

of the actual herbage intake of the animal. This technique employed in combination with the chromogen method of digestibility determination will be useful in evaluating the feeding value of forages under grazing conditions.

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